A TREATISE ON KALA-AZAR

IN PREPARATION.

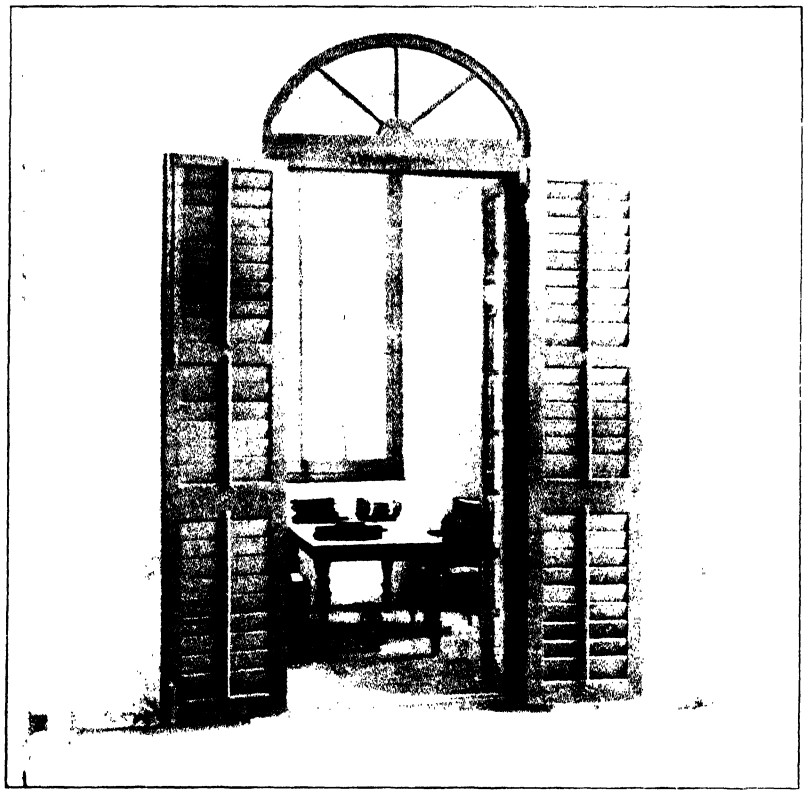
BLACKWATER FEVER

BY

U. N. BRAHMACHARI,

M.A., M.D., Ph.D., K.-I.-H. (Gold),

Formerly Additional Physician, Medical College Hospitals, Calcutta.



THE ROOM AT THE CAMPBELL HOSPITAL, CALCUTTA, WHERE
URIA STREAMINE WAS DISCOVERED BY
THE AUTHOR.

Here the new light dawned, shedding comfort, bringing relief, enlarging the bounds of knowledge, extending the frontiers of science. The light that will grow more and yet more resplendent with the progress of the years

A TREATISE ON KALA-AZAR

BY

UPENDRANATH BRAHMACHARI,

K.-I.-H. (Gold), Rai Bahadur, M.A., M.D., Ph.D.

ex-Physician, Medical College Hospitals, Calcutta, and On Kala-azar Treatment Research Enquiry, Indian Research Fund Association (1920-26). Formerly Physician and Teacher of Medicine, Campbell Medical School, Calcutta. Honorary Lecturer in Physiology in the Post-graduate Department and Member of the Board of Higher Studies in Physiology, University of Calcutta. Head of the Department of Biochemistry, University College of Science, Calcutta. Member of the Council of Tropical Medicine and Hygiene, International Congress of Medicine, London, 1913. Fellow of the Calcutta University. Fellow of the Asiatic Society of Bengal. Fellow of the Royal Society of Medicine. Examiner in Medicine to the University of Calcutta.

[Translated from the Author's Treatise on Kala-azar in German in Professor Dr. Carl Mense's Handbuch der Tropenkrankheiten, Vol. IV, 1926, thoroughly revised and considerably enlarged.

LONDON:

JOHN BALE, SONS & DANIELSSON, LTD.

83-91, GREAT TITCHFIELD STREET, W.1.

1928

DEDICATED

TO

MAJOR-GENERAL GODFREY TATE

M.B., K.H.S., I.M.S.

Surgeon-General with the Government of Bengal

AS A MARK

OF

THE AUTHOR'S GRATITUDE

PREFACE

THE Revised Second Edition of the author's book on KALA-AZAR AND ITS TREATMENT, which has now been exhausted, was published in 1925, and since then the Fourth Volume of Mense's HANDBUCH DER TROPEN KRANKHEITEN, in which the chapter on KALA-AZAR was contributed by the author, appeared in 1926.

The present volume, which is based upon this Chapter in Mense's work, is a new book, embodying all the latest facts known about the probable transmission of the disease by means of the sandfly, the description of "dermal leishmanoid," a new form of cutaneous leishmaniasis due to *Leishmania donovani* first discovered by the author, and the latest advances in the treatment of the disease by means of the aromatic antimonials, some of which were discovered by the author in the course of his researches. Infantile kala-azar, which was not dealt with previously in my book on KALA-AZAR AND ITS TREATMENT, has been incorporated in the present volume.

A new feature of the work is the inclusion of maps showing the geographical distribution of the disease. New plates showing the stages of development of the parasite of kala-azar in culture, sections of the spleen, liver and bone-marrow in kala-azar have been added. A summary of the classical works of Christophers, Shortt and Barraud on the morphology of the parasite of kala-azar and its life cycle in the sandfly has been given.

The author has attempted to give an exhaustive bibliography of the disease which, he hopes, will prove useful for purposes of reference.

Ten years have passed since the author warmly advocated the treatment of kala-azar with intravenous injection of metallic antimony. In the onward march of search after truth, an organic antimonial has been discovered by him which to-day stands pre-eminent in the treatment of the disease.

The author's grateful thanks are due to the Governing Body of the Indian Research Fund Association for their having placed at his disposal grants which enabled him to carry on researches into the chemotherapy of antimonial compounds in kala-azar infection and to discover a series of new antimonials possessing therapeutic value in the treatment of the disease.

He is under the deepest obligations to Major MURISON, I.M.S., Director of Public Health, Assam, for kindly allowing him to make use of his pamphlet regarding the preventive measures against kala-azar that have been recently adopted by the Government of Assam, and have already been fraught with brilliant results, and to Colonel KNOWLES, I.M.S., Professor of Protozoology,

Calcutta School of Tropical Medicine, for allowing him to freely make use of some of the manuscripts of his work on MEDICAL PROTOZOOLOGY before it was published, for his many valuable advices in the preparation of this work, for making cultural and inoculation experiments with the first recorded case of dermal leishmanoid, and for very kindly going through the proofs of his book. The author is also deeply indebted to him for his kind permission to reproduce in this treatise the plate on "Normal and Abnormal Blood Cells," from Knowles and Senior-White's MALARIA: ITS INVESTIGATION AND CONTROL.

His sincerest thanks are due to Lieut.-Colonel CHRISTOPHERS, F.R.S., C.I.E., I.M.S., Director, Central Research Institute, Kasauli, and formerly Director, Kala-azar Commission, for his very kindly going through a part of the chapter on the "Morphology and Life-Cycle of the Parasite of Indian Kala-azar," and to Major SHORTT, I.M.S., Director, Kala-azar Commission, for his very valuable help to the author in working out the histopathology of dermal leishmanoid and making photomicrographs of the sections of the tissues, and for many valuable suggestions.

The author is deeply indebted to Lieut.-Colonel BARNARD, C.I.E., C.B.E., I.M.S., Professor of Medicine, Calcutta Medical College, and to the other colleagues of the author in the Medical College Hospitals, Calcutta, for their kind courtesy in letting him study the clinical aspects of the disease in his own ward along with theirs; to Major SHANKS, I.M.S., Professor of Pathology, and the other members of the staff of the Department of Pathology, Calcutta Medical College; and especially to Dr. M. N. DE, of the same department, for the beautiful sections of tissues in cases of kala-azar and dermal leishmanoid, and for the kind help rendered by him to the author in working out their histopathology. He is also much indebted to Dr. BAINI PRASHAD, of the Zoological Survey of India, for some of the excellent photomicrographs, and to Mr. V. V. SAHONE, Meteorologist, Calcutta, for giving him the correct meteorological figures for different parts of India.

The author puts on record his indebtedness to the Librarian, Royal Society of Medicine, for his help in preparing the Bibliography, and to the Editors of the *Tropical Diseases Bulletin*, from which much help has been obtained in preparing the same, and to his own assistants, Dr. B. B. MAITY and Dr. S. C. BANERJEE, for the great help they have rendered in the collection of references; to Dr. B. B. SHAHA and Dr. S. P. MUKHERJEE, of the Carmichael Medical College, Calcutta, in going through the proofs, and to Dr. R. BANERJEA, of the National Medical Institute, Calcutta, for helping him in writing out the Chapter on Laboratory Methods.

No work dealing with a protozoal disease can be complete without frequent reference to the remarkable book on PROTOZOOLOGY, by C. M. WENYON, C.M.G., C.B.E., F.R.S., Director-in-Chief to the Wellcome Bureau of Scientific Research, and the author acknowledges the greatest help that he has received by frequent reference to this valuable masterpiece of this high authority in protozoology.

UPENDRANATH BRAHMACHARI.

82/3, Cornwallis Street,
Calcutta.

CONTENTS

LIST OF ILLUSTRATIONS

PLATES

ILLUSTRATIONS IN THE TEXT

KALA-AZAR

(INTERNAL OR VISCERAL LEISHMANIASIS)

CHAPTER I.

DEFINITION.

KALA-AZAR is a disease due to a general or systemic infection by a member of the leishmania group of parasites, which give rise to various affections included under the general name of leishmaniasis. It occurs in children and adults and is characterized by a high mortality in cases not treated with antimony, irregular fever, frequently running a chronic course, sometimes acute or subacute, and lasting from a few months to two or three years, or rarely more.

It is accompanied by enlargement of the spleen and frequently also of the liver, a gradual downhill course with progressive emaciation, anæmia, leucopenia leading to marked cachexia and hæmorrhage in different parts of the body. It is usually terminated by some intercurrent disease or cancrum oris.

SYNONYMS.

(Including Kala-azar of Adults and of Infants.)

Leishmaniasis, internal leishmaniasis, visceral leishmaniasis, general leishmaniasis, kala-azar of adults, tropical leishmaniasis, Indian kala-azar, black fever, black sickness, tropical splenomegaly, cachexial fever, cachectic fever, Leishman-Donovan disease, non-malarial remittent fever, malarial cachexia (in error), Dum-Dum fever, Burdwan fever, sirkari disease, sahib's disease, kala-dukh, kala-jwar, kala-hazar, Assam fever, tropical cachexia, tropical kala-azar, ponos (Greece), haplopinacon (Cephalonia), semieh (Sudan), infantile kala-azar (Nicolle), infantile leishmaniasis, Mediterranean kala-azar, Mediterranean leishmaniasis, febrile splenic anæmia (Fede), anæmia infantum a leishmania (Pianese), leishmania anæmia (Jemma and di Cristina), marda tal bicceia (Malta), malattia da mensa (Sicily), febrile pseudo-leukæmia infantum.

Broadly speaking leishmaniasis can be divided into the following clinical types :—

(1) Visceral leishmaniasis. This includes :—

(*a*) Kala-azar of adults, due to *Leishmania donovani*, Ross, 1903 (Laveran and Mesnil), occurring mainly in India.

(*b*) Infantile kala-azar, occurring especially in the Mediterranean countries, due to *Leishmania infantum* (Nicolle).

(*c*) Sudan leishmaniasis due to *Leishmania donovani*, var. *archibaldi* (Castellani).

The modern view is to regard all these parasites as one and the same, as will be seen later on.

(2) Dermal leishmanoid—a skin disease recently described by Brahmachari, to be found in persons cured of kala-azar by antimonial treatment. (See Chapter on Dermal Leishmanoid.)

(3) Local leishmaniasis. This includes :—

(*a*) Cutaneous leishmaniasis due to *Leishmania tropica* (oriental sore, Baghdad boil, Delhi boil, &c.) and cutaneous lesions due to *Leishmania donovani*.

(*b*) Indian oro-pharyngeal leishmaniasis described by Castellani.

(*c*) Leishmaniasis Americana (espundia, bubas Braziliana, uta, &c.), occurring in South America.

The name "kala-azar," though it has the sanction of usage, is not very appropriate. It has been stated that it was intended to signify a fever in which dark pigmentation of the skin is a constant symptom. Some of the observers consider that "kala-jwar" is the proper term, because "kala" means "black," and "jwar" means "fever." But "azar" means sickness, and the term black sickness is just as appropriate. The word "kala" may, however, signify *kal*, i.e., death, signifying a fatal illness. As was pointed out by Ross, the popular use of the adjective "kala" does not necessarily imply blackening of the skin. It may mean deadly, just as *kala-sarpa* means deadly snake. Similarly, as Ross pointed out, black death signifying plague indicated the terrifying effect of the disease on the imagination of the people, rather than the actual reality of the disorder. This explanation applies here also, because some cases of kala azar do not show great pigmentation of the skin.

HISTORY.

Rogers held that the old "Burdwan fever" (1854-75) was kala-azar. If this view is correct then the first epidemic manifestation of the disease could be traced to a peculiar type of fever occurring in Jessore in 1824 or 1825, called "Jwar-Vikar," which Elliot considered was very similar to "Burdwan fever." In 1882, Clarke first described the disease in his sanitary report of Assam, in which he pointed out that "there is a fever of malarial poisoning known among the natives as kala-azar or black sickness, from the darkened colour which the skin assumes in chronic cases." He also stated that "as far back as 1869, the attention of the administrative officers in Assam became directed to a peculiar disorder called kala-azar, the ravages of which decimated,

and in some instances depopulated, numerous districts in the Garo Hills." Clarke based his report on the records and notes of 120 cases by McNaught, Civil Medical Officer of Tura, the headquarters of the Garo Hills district. Rogers has stated that in the administrative reports of Assam for 1875, several families were stated to have died among the Garos of a disease called "kala-hazar." In 1889, Giles concluded from his investigations that the disease was ankylostomiasis, or rather a mixed anæmia brought about by ankylostomiasis acting on a population worn down by chronic malarial poisoning "similar to the beriberi of Ceylon." Dobson strongly opposed the ankylostoma theory. In 1884 Stephens stated in his yearly report that the disease was distinct from malaria though allied to it. In 1894 Hindley described the existence of a disease popularly termed "Pushkara," in Jalpaiguri, in Bengal, which was undoubtedly kala-azar.

In 1896 Rogers considered that kala-azar was a malignant form of malaria, and Ross looked upon it as malaria plus some secondary form of infection. In 1902 Bentley claimed that the disease was allied to Malta fever. Neil Campbell thought that it was ankylostomiasis plus malarial cachexia in 75 per cent. of cases, and in the remaining 25 per cent. either ankylostomiasis pure and simple or pure malarial cachexia. In 1898, Harold Brown investigated "kala-dukh" in the Purnea district, which, according to him, was allied to kala-azar of Assam. About the same year Ross investigated "kala-jwar" of the Darjeeling district, which was also a disease similar to kala-azar.

Manson believed that the disease was not malaria because of the absence of periodicity in the febrile attacks and non-amenability to quinine even when given in massive doses, and in 1903 he suggested that kala-azar might be caused by a trypanosome. A few months later in the same year, Leishman reported the discovery as early as 1900, of peculiar bodies in the spleen pulp of a soldier who died of Dum-Dum fever at Netley Hospital. He came to the conclusion that there was a similarity between these bodies and trypanosomes, and he published his conclusions in 1903 in a paper entitled "On the Possibility of the Occurrence of Trypanosomiasis in India," in which he described these bodies as degenerate trypanosomes. In July, 1903, Donovan reported the finding of similar bodies, independently of Leishman. He punctured, during life, the spleen of patients in Madras suffering from prolonged fever with splenomegaly. He contested Leishman's view that they were degenerate trypanosomes. Laveran and Mesnil, after examining the specimens sent by Donovan, concluded that the organisms were piroplasmata. Identical bodies were also found by Marchand in January, 1903, in sections of spleen, liver and bone-marrow from a Chinaman who had taken part in the Pekin campaign and had suffered from fever with enlargement of the spleen and anæmia, and died in a Hamburg hospital. A demonstration was given before the Leipzig Medical Society on February 3, 1903. In December, 1903, Manson found similar bodies during life in the spleen blood of a patient from Darjeeling, suffering from kala-azar, and showed that these bodies were not endo-corpuscular bodies, as had been supposed by Mesnil and Laveran.

In 1904 Christophers wrote his first report "On a Parasite found in

Persons suffering from Enlargement of the Spleen." In the same year Castellani discovered the parasites in kala-azar patients in Ceylon, and Bentley found these bodies in the spleen of cases of kala-azar in Assam. Christophers, after examination of the smears of spleen blood of cases of kala-azar of Assam, came to the conclusion that many cases of malarial cachexia and kala-azar of Assam were one and the same disease.

In the same year Rogers' paper, entitled, "Leishman-Donovan Bodies in Malarial Cachexia and Kala-azar," and his preliminary note on the development of the parasite of cachexial fever and kala-azar into a flagellate stage, were published, and it became obvious that kala-azar of Assam and many cases of so-called "malarial cachexia," which is prevalent in eastern parts of India, were one and the same disease. The term kala-azar quickly came to embrace these conditions. In 1906 Brahmachari wrote his paper, "On a Contribution to the Study of Fevers due to Leishman-Donovan Bodies."

In 1905 Statham published a preliminary note, "On the Cultivation of the Leishman Body." In the same year Leishman and Statham wrote a paper on "The Development of the Leishman Body in Cultivation."

Besides India, the first case in Asia was observed post mortem by Marchand in 1903. Soon afterwards cases were found by Bassett-Smith among sailors. In 1910 Aspland showed that the disease was widely spread in North China. In 1911 Cochran reported cases in other parts of China. Saville found it in Tientsin, and Jerusalémy found it in Hoang Ho district. Jeffreys and Maxwell found it in Formosa.

In Italy, the first clinical description of the disease was given in 1880 by Cardarelli. He was followed by Fede, Somma, and other Neapolitan workers, who thought that two clinical varieties of the disease existed. One of these was always accompanied by a rise of body temperature (*varietas febrilis*), while the second generally ran its course without producing fever (*varietas apyretica*). The second form was considered to include cases of anæmia splenica infantum pseudo-leukæmia (Henoch v. Jaksch). The febrile form was thought to be infectious. Fede, Pianese and Gianturco conducted bacteriological investigations, and the conclusion reached by Pianese and Gianturco was that the disease was caused by *Bacterium coli*. Mya and Trambusti found *Micrococcus tetragenus* present in the apyretic form of the disease.

In 1904 Neave discovered the existence of this disease in Africa in a child coming from Anglo-Egyptian Sudan, and in the same year Philips discovered it in two adults coming from Arabia. In the same year Cathoire observed peculiar bodies in smears from the spleen of a child who had died of an ill-defined disease in Tunisia, and they were subsequently recognized by Laveran as *Leishmania donovani*. In 1903 Laveran's paper on *Piroplasma donovani* appeared.

In 1905 Pianese found the identical picture in smears from the spleen and liver of children dying of infantile splenic anæmia in Italy. In 1907 Pirrie, who had been working in the Sudan, died in England of kala-azar. In 1908 Cummins discovered a case contracted at Sniga on the Blue Nile; Carrol

recorded a second case from the same district, while Black met with two cases; cases were then reported frequently from this district. Bousfield recorded seven cases from the province of Kassala and one from Mafaza. Thomson and Marshall found forty-one new cases in children and adults along the Blue Nile towards Abyssinia, which forms an endemic zone. Marshall observed the disease among children of 12 years of age in the Sennar province of Sudan. Archibald studied a small epidemic in Kurmok on the Abyssinian frontier, and the disease in the Sudan is sometimes called Sudan kala-azar. Nicolle and Cassuto in 1907 observed the parasites in the spleen of a child in Tunis suffering from irregular fever and splenomegaly. Nicolle named the disease infantile kala-azar.

From 1905 to 1908 Pianese observed only a few cases, and later on none at all were seen, so that in 1912 the disease was thought to have died out in Italy. Gabbi, however, reported the occurrence of the disease in Sicily and Calabria at about the end of 1908. During the years 1908 to 1914, Nicolle treated thirty-eight cases in Tunis.

From 1908 to 1913, 119 cases occurred in Messina, 100 in Catania, and 110 in Palermo. As a result of Gabbi's work, the problem began to be studied in Italy on a large scale. It appeared that kala-azar was fairly widespread in Calabria, in Sicily, and round Naples, and that isolated cases had also occurred in Fiumicino (Rome) and Trieste.

The disease was also observed in the Mediterranean islands. Archer in 1907 reported its occurrence in Cyprus, Critien and Babington in Malta (1910), Gabbi in Spetza (Grecian Archipelago, 1910). The latter was the first to establish definitely the identity of ponos and kala-azar, a fact that Gabbi, Mesnil, Laveran, Williamson and others had previously supposed.

In 1910 Tashinbbey discovered the disease in Tripoli, and in the same year Alvares discovered it in Lisbon. In 1911 Christomanos observed cases on the Greek mainland; it had already been observed at an earlier date on the archipelago by Aravandinos and Michaelidis. Since 1912 cases have been described in Russia (Taschkent, Turkestan) by Marzinowsky, Petroff, Nikoforoff, Gourko, Yakimoff, and others. Pittaluga and others observed cases in Spain in the same years. The disease has also been found in Moscow.

The reader may consult the list of references in the introductory number of *Kala-azar Bulletin* (1911) for further references regarding the history of the disease.

GEOGRAPHICAL DISTRIBUTION.

Asia.—The disease occurs extensively in the eastern parts of India, especially in the districts through which the rivers Ganges and Brahmaputra pass. There is also an endemic area in Madras, from which many cases originate. The provinces of Assam, Bengal, parts of Madras, Behar and Orissa, and the eastern parts of the United Provinces are the chief endemic

areas (see Plates I and IV). A new endemic area has been discovered in the south of the Madras Presidency. Childe mentions the case of a European lady who contracted the disease either in Bombay or Jubbulpur. Another case has been recently reported on the west coast of India. It has been observed in Ceylon, where it is rare, and cases have been reported in Burma as well as in Siam, Sumatra and Formosa. There are endemic areas in certain parts of China, such as Pekin, Central China, and the district of Yang-tse-Kiang, and more recently in some parts of South China. The disease occurs also in Asiatic Russia, Russian Turkestan, Arabia and Asia Minor. It is doubtful whether it occurs in Mesopotamia, though Külz considers the disease to be endemic there.

Europe.—The disease is endemic in many parts of Europe bordering on the Mediterranean, e.g., the southern parts of Italy and in Sicily, and especially in Calabria and the neighbourhood of Messina. In Messina the first villages wherein the disease was recognized were Bordonaro, Camaro and Galati (Gabbi). Other endemic areas were discovered later, e.g., Catania (Felitti and Longo), Palermo and the neighbouring country (Jemma), Naples and the surrounding districts (Petrone, Vaglio and Gabbi). Recently, a case of adult kala-azar has been observed in Madrid (see Plate II).

Cases have also occurred in Trieste. In Malta, too, the disease is endemic. It occurs in Greece, chiefly in the islands of Spetsa and Hydra. In Hydra, the annual death-rate from the disease is estimated at 7-10 per 6,000 inhabitants. In 1911, fourteen children (39 per cent. of the death-rate) died of kala-azar. Isolated cases of the disease have been reported from southern Spain and Portugal and from France in Marseilles and Nizza. Cases have also been described in Russia (Moscow, Mohileff) as well as in Vienna and Riga.

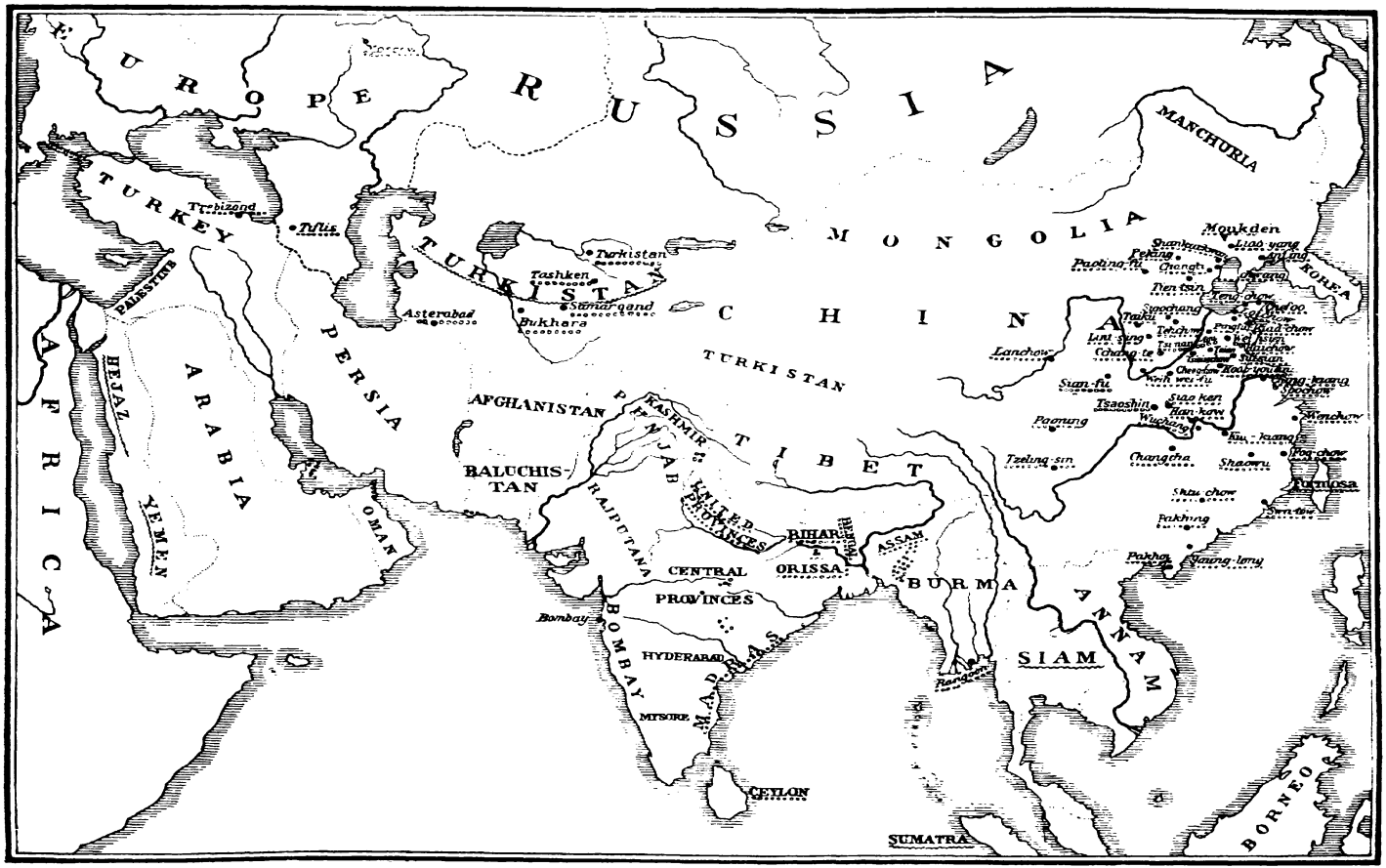
Possibly anæmia splenomegalica, which Seyfarth observed in the coastal district of southern Bulgaria, was some form of kala-azar. In Macedonia cases may also be found.

America.—One case has been observed in South America by Migone (?).

Africa.—It is endemic in almost the whole of the Mediterranean coast. It is said to be exceedingly rare in Egypt (Archibald). Gabbi states that it occurs in Egypt. It is endemic in Tunis, Tripoli, Algeria, Cyrenaica, Jat Lakes, Morocco. There is a limited endemicity in the Kassala and the Blue Nile districts adjoining the Abyssinian border. Endemic centres possibly exist in Eastern Sudan and Western Abyssinia. One case occurred in Khartoum (Gabbi), one in the Jehad territory and one in Gabon. With the exception of one case described by Bouillez in central Shari, the disease has hitherto not been found in the French, Belgian, former German, Spanish and Portuguese possessions in West and Central Africa, nor in South-west or East Africa—(see Plates II and III).

The first endemic area discovered in Africa was Tunis. Cases of kala-azar have recently been observed in Egypt, in Cyrenaica (Patane), near Lake Chad (Bouillez), in East Africa (Lafont, Heckenroth), and in Morocco (Klippel and Monier Vinard)—see Plates II and III.

The disease has been discovered in Kenya Colony (Wenyon).



oooooooo = Endemic area ~~~~~ = Area possibly infected

DISTRIBUTION OF KALA AZAR IN ASIA.

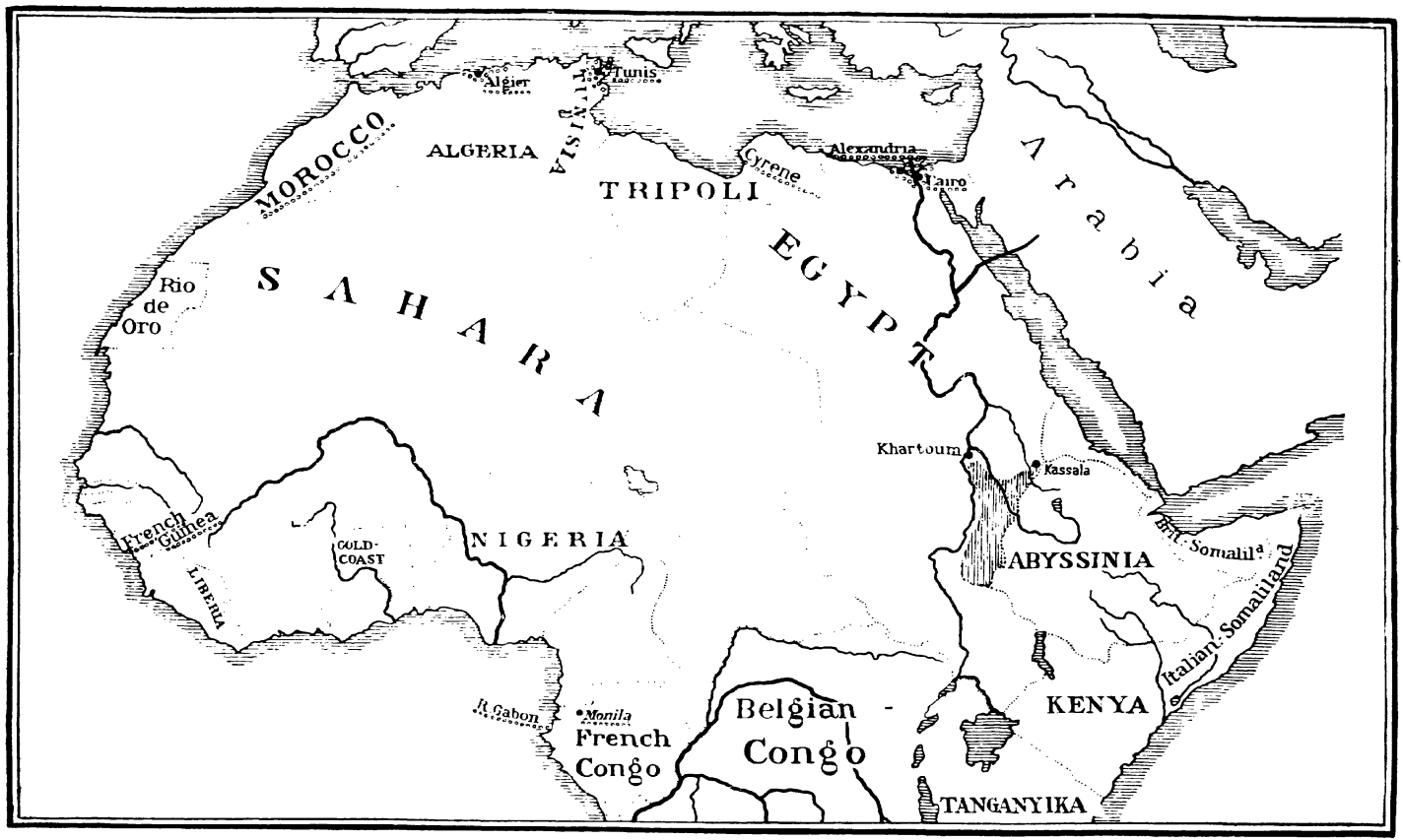
(Original.)

PLATE I.

⁘⁘⁘ = Areas of scattered cases and endemic centres.

DISTRIBUTION OF KALA-AZAR IN THE MEDITERRANEAN COUNTRIES.

(Original.)



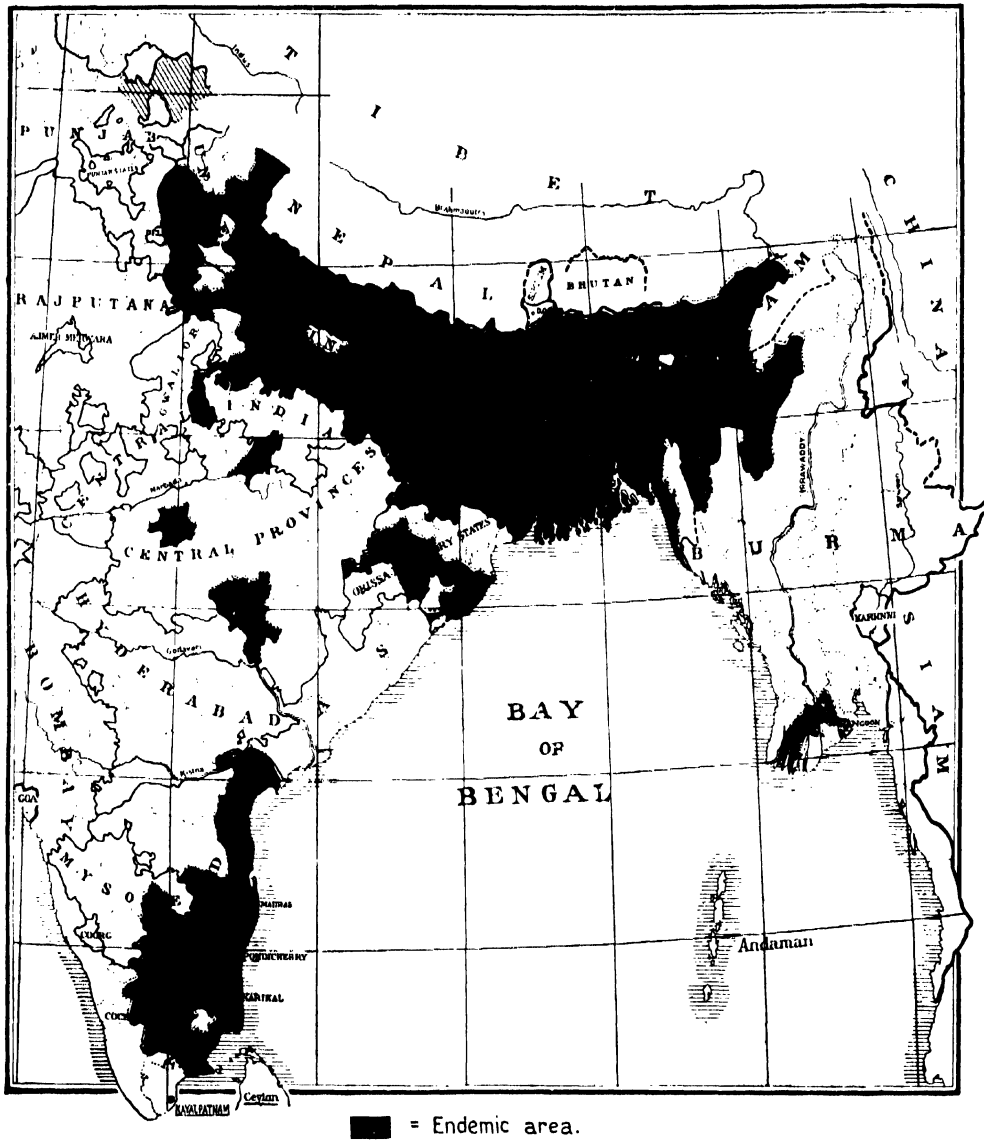
▒▒▒ = Areas of scattered cases and endemic centres. ▓▓▓ = Endemic area on the Upper Nile.

DISTRIBUTION OF KALA-AZAR IN AFRICA.

(Original.)

PLATE III.

PLATE IV.



DISTRIBUTION OF KALA-AZAR IN BRITISH INDIA.

It is impossible to give a complete list of all the places in the world where kala-azar might be found. Reference should be made to the accompanying Plates I to IV for such information.

EPIDEMIOLOGY.

In India, epidemics of kala-azar have only been described in certain parts of Assam and Bengal. On the other hand, the disease is endemic in the other parts of India which it infects. These include the provinces of Madras, Behar and Orissa, and the eastern parts of the United Provinces of Agra and Oudh.

In Bengal, Rogers held that the old Burdwan fever (1854-1875) was an epidemic of kala-azar. Subsequently Brahmachari concluded, both from clinical and statistical evidence, that the old Burdwan fever was mostly of malarial origin. In more recent times epidemics have been described in Assam, in the districts of Purnea (Brown), Darjeeling Terai (Ross). It is in Assam that epidemics occurred up to recent times.

The epidemic fevers of Dinajpur and Rungpur in Bengal which occurred, according to Rogers, between 1871 and 1876, but which probably also occurred before the seventies, were thought by him to be epidemic manifestations of kala-azar. According to him the epidemic fevers of these districts, the epidemic of kala-azar of Assam and the kala-dukh of Purnea were manifestations of one and the same disease. He thought that the disease was started in Rungpur district in the early seventies by the extraordinary succession of unhealthy seasons, and that it spread slowly up to the Assam valley, giving rise to the epidemic of kala-azar in Assam. There was an epidemic fever in the district of Patna (Behar and Orissa) in 1850-59, which somewhat resembled the old Burdwan fever.

THE ASSAM EPIDEMIC.

It is practically certain that Assam is the part of India where epidemics of kala-azar once acquired a home. The first epidemic in Assam is supposed to have commenced in the Garo Hills about 1870. It spread steadily from year to year in an easterly direction along the Brahmaputra valley, following lines and roads of communications, and continuing for about ten years, and then slowly declining. Recently there have been other epidemics in Assam.

The nature of the epidemics of kala-azar is characterized by certain features :—

(1) The slowly spreading nature of the epidemic, travelling at the rate of ten miles a year and always along the routes of traffic.

(2) When it attacks a particular village, it clings to particular houses, and then spreads from one house to another by means of communication.

(3) The disease is not diffused generally over a district, but settles in small foci.

The chief factors in its spread appear to be :—

(*a*) Human carriers of the disease.

(*b*) Close association of the sick with the healthy.

(*c*) Probably the presence of a blood-sucking insect as the intermediate host.

(*d*) Climate and local conditions of the soil compatible with the rapid spread of the disease.

(*e*) Absence of any immunity in the community.

The investigations of the epidemiology of kala-azar in India have suggested a number of problems which have hitherto remained unsolved. Why did the epidemic spread slowly from one place to another, instead of extending rapidly over a whole region? Why did epidemics occur only in Assam, and not in other parts of India or elsewhere where the disease is endemic? The suggestion that has been made, that epidemics occur when the disease spreads to a virgin soil, is not satisfactory, as it has not been proved that epidemics have ever occurred in parts of India where the disease is now endemic, except Assam and perhaps Bengal. It may, however, be assumed with considerable probability, that in an infected endemic area, there may be a great increase in the percentage of deaths among the infected people due to unhealthy conditions developing in a particular season or year. An epidemic of another fever such as malaria or influenza may have attacked those previously suffering from endemic kala-azar, and suddenly raised the mortality curve of the disease, thereby giving it an epidemic character.

It may be pointed out here that endemicity forms a special feature of kala-azar. Donovan doubted the possibility of an epidemic outburst in a disease of the nature of kala-azar. It is possible that in some of the epidemics malaria and kala-azar ran together, and that in some of them one disease was more prevalent than the other. Perhaps in the Assam epidemic in 1870 kala-azar was more common, and in the Burdwan epidemic (1854-75) malaria was more common.

In Tunis, Algeria and Portugal, there has never been an epidemic of kala-azar. Gabbi has collected information in order to discover at what time of year most of the cases occur. He found in Sicily that the disease first appeared in spring, and that most cases occurred in April and May. In June and July there was some diminution in the number, and by August, September, October and November, the disease had completely or almost completely disappeared. The curve representing the course of the disease reaches its highest point in the month of May. It must certainly be added that epidemics, in the true sense of the word, have never been observed in Mediterranean countries. Archibald described a small epidemic occurring in Kurmok, in the Sudan, on the Abyssinian border, and recently such an epidemic has been reported from China.

CHAPTER II.

ÆTIOLOGY.

PREDISPOSING CAUSES.

KALA-AZAR is generally a sporadic disease and its endemicity has very special characteristics. The possibility of the existence of chronic carriers of the disease in endemic centres should be borne in mind. Patients with anæmia and œdema, but without enlargement of the spleen and liver and without fever have been observed in these places. They harbour leishmania bodies in their blood and these may be carriers of the disease.

It has been found in Mediterranean countries that the disease occurs during or immediately after the period of dentition (Gabbi, Laveran).

It has often been observed in children, following on an attack of exanthematous diseases, such as measles or scarlet fever (Gabbi, Laveran).

There are various conditions which have a bearing upon the spread of infection :—

(1) *Domestic Conditions.*—In the endemic areas of India the disease has a tendency to run in families living in the same house. Indeed, it may be said that the disease tends to cling to the same household. In Bengal it does not always affect individuals living in the same house at one and the same time ; the husband may be attacked one year, and the wife some years later. In Assam and sometimes in Bengal, several individuals may, on the other hand, be found affected at one and the same time.

Similarly in Italy and Sicily, though several attacks have been observed in the same family, yet children in the same family living under similar surroundings and hygienic conditions and even in the same bed, are very rarely or never attacked at one and the same time. This applies also to the other Mediterranean countries. In the Sudan it is also exceptional to find more than one case in one family or in one household. The same may also happen among the sporadic cases in the affected parts of Bengal, where only one member of the family may be attacked, the others escaping infection. It may sometimes be found that a servant living in the same house has suffered from the disease previously. Some observers in Calcutta have found that only a small proportion of cases give a history of another occupant of the same house having the same disease. In some cases, however, multiple infections have definitely occurred in Calcutta.

(2) *Age.*—The age incidence is somewhat variable in different parts of the world. In the Mediterranean countries, and certain provinces of China, it is mostly infants that are attacked ; however, cases of adults contracting the

disease have also been reported from Malta, Cyprus, Sicily, Greece and Russia. (Archer, Philips, Babington, Bassett-Smith, Christomanos, Gabbi, Gourko, Petrow and others.) In the Egyptian Sudan older children and early adolescents are somewhat more frequently attacked than adults. In the region of the Caspian Sea children and adolescents are alike attacked. Some of the available statistics in India and China are as follows :—

In Mackie's statistics, out of 195 cases, 9 were 1 to 5 years of age, 100 were 6 to 10, 49 were 10 to 15, 17 were 16 to 20, 12 were 20 to 30, and 8 were over 31, that is 76 per cent. were between the ages of 6 and 15 years. In Rogers' statistics in his Assam series, 25·6 per cent. were under 10, and 24·4 per cent. between 10 and 20 years of age. Among the sporadic cases of Indians in Calcutta, 8 per cent. were under 10, 40 per cent. between 10 and 20, 32 per cent. between 20 and 30, 16 per cent. between 30 and 40 years of age, and only 4 per cent. above 40 years of age. In China, Cochrane found 32·6 per cent. between 1 and 10, 29·6 per cent. between 21 and 30, 21·6 per cent. between 31 and 40, and only 3·6 per cent. between 41 and 50 years of age. In a series of hospital cases collected by me some years ago, nearly 33 per cent. of the cases were under 20 years of age. In a subsequent series of 118 cases in the same hospital, 3 were under 6, 20 between 7 and 12, 39 between 13 and 20, 38 between 21 and 30, and 18 above 30 years of age, that is, nearly 85 per cent. were under 30 years of age. Other statistics show that 50 per cent. of the cases are under 20, and that most of the cases were between 3 and 10 years of age. According to McCombie Young, kala-azar is also very widespread among children in Assam. The hospital statistics do not give an accurate idea of the number of children affected, and in consultation practice one meets with a much larger percentage of cases in children under 6 and some even less than 1 year old. The lowest age at which the disease was observed by me was 6 months.

During epidemics the disease probably attacks persons of various ages in proportion to their age distribution in the population. In Assam the proportion of children affected is said to be greater than in Bengal, but this requires further investigation. It is quite certain that adults may be attacked in places where infantile kala-azar is most common, and *vice versa*.

(3) *Sex.*—In Calcutta the number of males admitted into hospitals is greater than that of females and, according to Rogers, in proportion to those of other diseases. In his statistics, among Europeans there were twice as many males as females above the age of 15, but in children under 15 there were twenty-three females to thirteen males. In one of my series the number of females admitted into the Calcutta Campbell Hospital was less than could be accounted for by the proportion of females admitted for treatment for other diseases. This was in part due to the fact that this series contained only those which gave positive results on spleen puncture, and this operation was performed less frequently on female patients. In consultation practice the number of affected females is much less than that of males. Thus out of a series of 120 cases observed by me there were ninety males and thirty females. Rogers found that among tea-garden coolies there was no difference in

the incidence of the disease between the two sexes. In China, Cochran saw only four females among eighty-three cases.

In Italy statistics vary on this point. Gabbi found women preponderating among his cases. According to Longe, Jemma and his pupils, there is no particular difference. In the Sudan, females are attacked only exceptionally.

(4) *Race.*—No race is exempt from this disease. In a series of 118 cases observed by me in the Calcutta Campbell Hospital, there were ninety-eight Hindus and twenty Mahomedans, which would make the proportion of Hindus to Mahomedans five to one, and this was found to be the proportion of total number of Hindus to Mahomedans admitted to the hospital. In Assam, Mackie found that the Jain community was free from this disease in the affected areas. Later on, however, it was observed among members of the Jain community who came from affected areas of Assam and elsewhere. In endemic areas the coloured races are chiefly affected, and although the white races are not exempt, no epidemic has yet been observed among them.

British soldiers in Dum-Dum camps were formerly affected, hence the name "Dum-Dum" fever.

Better class Europeans and Anglo-Indians are much less affected in Bengal than the poorer classes. In Assam, the English tea-planters are more frequently attacked than the Europeans in Bengal. The poorer classes of Indian Christians in Calcutta are frequent sufferers from kala-azar. A British high official, who had the disease in the United Provinces, came under my observation.

(5) *Climate and Seasonal Incidence.*—Kala-azar occurs only in warm countries (tropical and subtropical). In those parts of India where it occurs the year may be divided into three periods, according to variations in the rainfall: the cold season from October to March, the hot season from March to June, and the rains from June to October. At the beginning of the cold season medium temperatures prevail generally. These continue falling till January, except in the Peninsula, where December is the coldest month, and then begin to rise. In Madras the average temperature during the months of December, January, February and March may range between 75° F. and 85° F.

The magnitude of the annual variation of mean temperature in India is roughly as follows: about 35° F. in north-west India, about 15° F. in the Peninsula, and about 20° F. in the other parts of India. In Calcutta the temperature ranges between a monthly mean maximum of 96° F. (April) and a monthly mean minimum of 56° F. (January). In Madras the corresponding values are 99° F. (June), and 68° F. (January). In parts of the country some distance from the coast the range is naturally greater.

The disease is found in districts which have a mild winter (60° F. to 75° F.), and, according to Rogers, cases occur during this season in proportion to the length of time that the cool weather lasts. It is found inland and in mountainous districts, as well as along the coast.

The rainy season, which is brought by the monsoon wind, begins in the last week of May, in Ceylon, Burma, and Southern India. In Central India,

Bengal and the West Central Provinces, it begins during the second week of June, and in the north towards the end of June. The monsoon lasts until September, and in October the rains cease. It is during this period that fewest cases of kala-azar occur, according to Rogers.

In my experience new cases came to Campbell Hospital in Calcutta for admission throughout the year. The largest number of admissions into my wards in that hospital were from June to October. In Rogers' statistics there were three to four times as many cases from November to April as in the remaining six months of the year. According to Dodds Price, every one of his cases in Assam commenced during the cold weather. In the Sudan the disease appears to be more prevalent in the period following the rains, i.e., between August and December.

In considering the seasonal incidence of kala-azar, the long incubation period of the disease has to be borne in mind.

In Mediterranean countries the disease occurs chiefly at the end of the winter and in the spring (last period of the rainy season) (Gabbi).

The average temperatures in Sicily from November to May are given below :—

November	...	...	...	...	58° F.
December	...	...	...	...	48·4° F.
January	...	...	...	...	49·4° F.
February	...	...	...	...	48·7° F.
March	...	...	...	...	52·3° F.
April ...	...	...	...	...	57·7° F.
May ...	...	...	...	...	65·8° F.

Syracuse, Tripoli, Trapani and Tunis have approximately the same climate.

In the Mediterranean countries east and south-east winds predominate in autumn, winter and spring, and north and north-west winds in summer.

In the Mediterranean countries the disease has a well recognized predilection for sea ports, which are connected with land on the east side.

McCombie Young considers that humidity rather than temperature is the chief determining factor in the distribution of kala-azar, but temperature, especially minimum temperature, is also of some importance, and the conditions of temperature and humidity required for the transmission of infection are as follows :—

(1) A considerable degree of humidity, not less than is indicated by an annual mean relative humidity of 60 per cent.

(2) A monthly mean minimum relative humidity which is not at any time less than 40 to 50 per cent.

(3) A mean monthly minimum temperature not falling lower than 50 F.

(6) *Habitation and Position.*—Up to what height above sea level have cases of kala-azar been observed ? Exact information on this point does not exist. In Sicily, Signer observed cases in Marinco 613 metres above sea level, and in Salemi 442 metres above it. The opinion that the disease does not occur at heights over 2,000 ft. (600 metres) above sea level requires further

evidence in its support. Cases have been reported to the author from higher altitudes, but it cannot be definitely stated where the disease was contracted by them. Recently Savage has observed two primary cases of kala-azar in Sanawar, in the Simla Hills, at a height of 5,760 ft. above sea-level (1927).

In the Mediterranean countries the disease is more prevalent in districts near the sea. In towns, the outskirts and rural districts are most affected. In the Sudan the disease occurs in villages adjacent to khors and rivers (Archibald, Bousfield, Thomson and Marshall). In India the disease occurs more frequently in villages than in towns, except in the city of Madras, where there is a definite endemic area. Sporadic cases have been observed in which the infection could be traced to Calcutta. The disease was contracted in dilapidated, ill-ventilated, overcrowded houses, or insanitary bustees, or in houses adjoining them. In one or two places, a number of houses near to a particular tank or pond were infected more than others. In India, a more or less low-lying district with a damp climate and a heavy rainfall, in the neighbourhood of stagnant or running water, seems favourable for the progress of the disease. In Italy the infected houses are generally overcrowded, ill-ventilated, and occupied by domestic animals, especially dogs.

Archibald has shown that in the Sudan the disease occurs in characteristic, limited, endemic areas. The researches of Laveran, Franchini, Fantham and Porter suggest the possibility of the distribution of the disease being in some way connected with the presence of cysts of the herpetomonads of certain arthropods in the water.

(7) *Occupation and Social Position.*—The disease is specially common among the poorer classes, such as workmen and agricultural labourers, but it is also not uncommon among the middle classes in India. In Italy it is found more frequently among workmen and peasants.

Napier has given a list of complicated factors which he considers play a part in determining the spread of the disease by means of a hypothetical transmitter. They raise an extremely complex and perplexing problem. Some of the factors supposed to have a bearing on the incidence of the disease are different in towns as compared with rural districts. Others, such as an altitude of less than 2,000 ft. above the sea-level, or keeping of chickens or ducks in houses, do not appear to be essential factors, and, as has been pointed out by Charles Young, it is doubtful how far some of the data formulated by Napier are relevant to the question of infection. The Jain and Marwari community, who never keep ducks or chickens in their houses, are not immune.

EXPLANATION OF THE DIAGRAMS ON PLATE V.

1. Mononuclear cell from the spleen with Leishman-Donovan bodies.

2-3. Smears of spleen pulp, containing various forms of the parasite, including small torpedo forms.

4. A later stage in the development, showing cell division in progress.

5. Group of young parasites. Below, some flagellate forms.

6. Small group of oval forms; none flagellate.

7. Young flagellate forms.

8. First stages in cell division of flagellate forms.

9. Further progress of cell division.

10. Mature flagellates with dividing forms.

11. Group of flagellates.

12. Rosette formation.

13 and 13*a*. Mature flagellates with chromatin granules

14-16. Mature flagellates.

17. Flagellate from which spirillar forms are forming (Leishman).

17*a*. Spirillar form (Leishman).

18. Polymorphonuclear leucocyte containing Leishman-Donovan body.

19. Cell of spleen pulp containing many Leishman-Donovan bodies.

20. Endothelial cell containing a large number of Leishman-Donovan bodies.

PLATE V.

Stages of development of *L. donovani* in culture. (Modified from Leishman and Statham.)

CHAPTER III.

ÆTIOLOGY (*continued*).

THE CAUSAL MICRO-ORGANISM.

THE history of the development of our knowledge of the causation of the disease has already been described.

The parasite of kala-azar is similar to bodies found in oriental sore, by Cunningham in 1885, by Firth in 1891 and by Wright, of Boston, in the tissues of tropical ulcer in 1903. The parasite occurs in the tissues of man as a non-flagellated body, and Rogers was the first to point out that when cultured under special conditions it could develop into a flagellated stage.

It was originally supposed that the parasite found in patients suffering from kala-azar in India was different from that found in infants in the Mediterranean countries, and accordingly two diseases were described :—

(1) Indian kala-azar.

(2) Infantile kala-azar of the Mediterranean basin.

The former was supposed to be due to *Leishmania donovani* and the latter to *Leishmania infantum*.

The fact that the disease is prevalent among infants, especially in the Mediterranean districts, and among adults in India, and the supposed relationship between infantile kala-azar and canine leishmaniasis, supported this view. But in more recent years it has been proved that adults also suffer in the Mediterranean countries. Similarly children under the age of one year have been attacked in India. The modern view is that of Gabbi, Wenyon and others, who hold that all forms of kala-azar are due to one and the same parasite. But this does not explain why the incidence of the disease is greater among infants than among adults in one part of the world, and *vice versa*. This fact still remains unexplained.

ZOOLOGICAL POSITION, MORPHOLOGY, AND DEVELOPMENT OF THE PARASITES.

The parasite was considered by Ross to belong to a special genus *Leishmania* (1903). Rogers, and subsequently Patton, thought that it belonged to the genus *Herpetomonas*. The name *Herpetomonas donovani* was suggested by Laveran and Mesnil (1903). Other classifications now abandoned were, *Monadidæ* (Cunningham, 1885), *Piroplasma* (Laveran and

Mesnil, 1903), *Helcosoma* (Wright, 1903), *Microsporidium* (Christophers, 1904), *Trypanosoma* (Leishman, 1903).

Recently Christophers, Shortt and Barraud have still adopted the name *Herpetomonas donovani* (1926). Wenyon, however, considers that the inclusion of *Leishmania* in the genus *Herpetomonas* cannot be admitted, as the members of the genus *Herpetomonas* have definite trypanosome stages which do not occur in *Leishmania* (PROTOZOOLOGY, 1926).

MORPHOLOGY AND DEVELOPMENT.

The Leishman-Donovan body is a small ovoid or rounded organism. The size of the parasite varies considerably. When spherical, it measures from 1 to 3 microns in diameter. More usually it is ovoid, with the long diameter measuring 2 to 5 microns and the shorter 1·5 to 2·5 microns. Sometimes it presents torpedo-like forms, especially when blood and not pulp is drawn by spleen puncture. It consists of a mass of cytoplasm covered by a definite membrane. On staining with Giemsa's stain after treating in methyl alcohol, it shows two chromatin masses, one larger than the other, enclosed in a mass of cytoplasm. The larger chromatin mass is the nucleus or macro- or tropho-nucleus, and the smaller one is the micro- or kinetonucleus, or kinetoplast. The former is a more or less spherical body lying against the membrane and is somewhat flattened on this side. The kinetoplast is usually rod-shaped, and is either at right angles, or at a tangent to the macro-nucleus. It stains more deeply than the latter. It may also appear as a point. One or more vacuoles may be present in the finely granular cytoplasm.

In deeply stained parasites a red line can be traced from the blepharoplast, which lies near the centre of the kinetoplast, to the surface of the parasite. This is the axoneme which give rise to the flagellum in culture.

Multiplication.—The parasite multiplies by simple fission, which is always preceded by elongation, and the division of the macro- and micronuclei, and the plane of division is always longitudinal.

Some forms, having attained a larger size, apparently appear to undergo a process of multiple division. These are almost circular, and have as a rule six chromatin masses, three large and three small, the former being placed round the periphery. According to Wenyon, the bodies showing multiple segmentation probably represent detached portions of cytoplasm of the large cells containing leishmania.

The parasites, as they occur in man, are probably almost invariably intracellular. They grow and multiply within the host cell, causing it to enlarge and then to disrupt after disintegration of the nucleus. After being set free the parasites are taken up by the endothelial or the polymorphonuclear or large mononuclear leucocytes. In smears they are often seen free, or in clusters of various numbers, sometimes arranged with great regularity. Sometimes as many as 50 to 200 or even more parasites are found together, embedded in a structureless matrix or stroma, probably the remains of the original host cell.

The following description of the development of the parasites is mostly taken from that given by Leishman (see Plate V) :—

In his observations the bodies obtained by splenic and liver puncture were incubated under aseptic conditions in test tubes, each containing 1 c.c. of a 4 per cent. sodium citrate solution, at a temperature of 17° C. to 22° C.

After two days the little oval parasites begin to grow, and their protoplasm takes on a deeper blue stain, becoming opaque and more granular. Then they commence to enlarge but retain their original shape. This initial enlargement with the retention of the original shape is due in part to the enlargement of the macronucleus, which is followed by an increase in the quantity of protoplasm, which now stains blue more deeply than before. The micronucleus appears somewhat enlarged, but retains its original characteristic shape. A marked feature in the development is the early appearance of vacuoles in the protoplasm, which persist in all stages of development (fig. 1). These were not observed by Rogers. Growth at this stage is very rapid, the protoplasm and macronucleus both continuing to increase. The chromatin

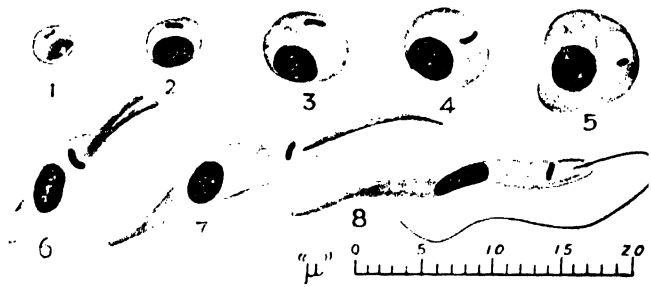


FIG. 1. Appearance of vacuoles in Leishman-Donovan bodies during development (Nos. 4, 5). Development of flagellum (Nos. 6-8). (After Leishman and Statham.)

becomes loosened and stains less deeply, but there is no further alteration of the micronucleus at this stage.

From the earliest stages even in very young cultures, multiplication by fission occurs. The first sign of this fission is seen in the macronucleus, which becomes successively elongated, constricted in the middle and finally separated into two distinct parts. Similar division of the micronucleus follows. This may, however, sometimes precede the division of the macronucleus. Schulz maintains that the nucleus divides by mitosis. The process of fission is completed by simple cleavage of the parasites into two "primary parasites." Sometimes one may give rise to three or even four new individuals by a process of further subdivision of the macronuclei and micronuclei. Many of these developmental changes up to and even beyond this stage may take place while the parasites are still embedded in the protoplasm of the splenic macrophages. Christophers also described secondary fission forms, composed of bodies of varied sizes, as contrasted with primary fission forms.

The flagellated stage rarely appears before the third day, but may occur within twenty-four hours.

The first stage towards flagellate formation is an alteration in the shape of the parasites. They become elongated, being usually thicker at one end than the other, and often distinctly pyriform. Sometimes flagellation may take place while the parasite is still definitely circular in form. Flagellation is a very rapid process. At first there appears a circular pink-staining area in contact with one side of the micronucleus, which may sometimes be more diffuse and merge in the blue-staining protoplasm of the parasite. Sometimes this pink zone may attain a size equal to half the diameter of the parasite itself. It has been termed the "flagellar vacuole." Following on this enlargement of the vacuole, there is rupture of the thin rim of protoplasm which forms the external border of the parasite, and protrusion of part of the contents of the vacuole in the form of a fringed process, or tuft, of the same staining reaction as the contents of the vacuole, and also of a fully-developed flagellum, and having the appearance of a bunch of pale pink threads, emerging from the neighbourhood of the micronucleus, passing onwards through the partially collapsed flagellar vacuole, and projecting clear of the body of the parasite. The filaments united and formed the rudiments of the flagellum. Wenyon considers that these appearances are probably artefacts due to a rupture of the vacuole, and that the axoneme, which is sometimes visible in the leishmania, continues its growth and extends through the surface of the parasite to form the flagellum.

In the next stage, the flagellum tapers slightly from its origin at the bottom of their flagellar vacuole towards its free extremity, while its length is now half that of the parasite. Then it grows rapidly. In some of the fully-developed parasites the flagellum appears to be inserted directly into the protoplasm of the parasite, near to, but not directly connected with, the micronucleus. The flagellum always arises from the rounded end of the parasites, and projects at once clear of the body. When the parasites move they advance with the flagellar end foremost.

A characteristic feature of the cultures is that the flagellates tend to remain clustered in groups, with their flagella directed towards one another, so that rosettes or spheres of organisms are formed with the flagella entangled at the centre. Wenyon considers that the fully-formed flagellate is flattened like a blade of grass. Sometimes one edge of the organism is convex and the other slightly concave, giving it the shape of a curved sword blade.

Considerable variations in the shape and structure of the macronucleus are found. It is doubtful whether these are due to a sexual differentiation among the flagellated forms. The macronucleus is generally granular, and in many cases there is a wreath or ring of chromatin granules enclosing a non-granular central zone. These granules are most probably chromosomes.

In many of the parasites small chromatin dots, distinct from the macro- and micronucleus, are found in the protoplasm. They tend to occur in pairs, and resemble small diplococci.

Multiplication by longitudinal fission occurs during the flagellated as well as during the non-flagellated stage.

A curious process of unequal longitudinal fission has been observed in some of the mature flagellate forms, and in this process the nuclei of the parent did not appear to play any part. The result of this process was the splitting off of a spirillum-like parasite from one end of the mature parasite. When newly separated, they are destitute of a flagellum, but it is subsequently developed, and these forms are very motile. Leishman considers that it is possible that these may undergo a further process of longitudinal division, giving rise to ultra-microscopic bodies (Plate V, 17, 17a).

The flagellates possess no undulating membrane, as do trypanosomes.

No process of the nature of conjugation was observed by Leishman. The body of the fully-developed flagellate is 14 microns to 20 microns long, and in the average 2 microns broad, while the flagellum is 16 microns to 24 microns long.

The following is the most recent description of the morphology and life cycle of *Leishmania donovani*, as given by Christophers, Shortt and Barraud.

The usual length of the body of the parasite, measured in wet fixed films, varies from about 9 to 16 microns, but stumpy forms may be as short as 5 microns. The breadth is usually about 1·5 microns, but may reach 2 microns or more in stumpy forms. The broad forms are often preparatory to, or actually in process of division, but broad forms giving no evidence of division occur. The length of the flagellum is frequently about the same length as the body and rarely exceeds twice this, except in globular or subglobular forms, where it may exceed four times the body length. (Christophers, Shortt and Barraud.)

ORIENTATION.

The Leishman-Donovan body, as seen in spleen smears, is a globular or ovoid parasite, having near the surface dorsally a kinetoplast with a parabasal from which a rhizoplast extends anteriorly, and ventrally a trophonucleus.

The trophonucleus and parabasal are situated at opposite poles on the periphery of the parasite and the rhizoplast is peripherally situated and always runs in a direction at right angles to the long axis of the parabasal.

The trophonucleus often shows an indication of being double. The arrangement of the parts may vary, the divided portions often being more or less anteriorly and posteriorly situated, though they are frequently seen symmetrically arranged on either side of the sagittal plane.

The rhizoplast is a linear structure about half the diameter of the parasite in length. It may appear straight, but is more frequently seen as a somewhat curved raphé-like line. It is probably at, or very close to, the surface whatever its position may appear to be.

The flagellar vacuole lies deeply situated between the rhizoplast and the trophonucleus, more or less in the hollow of the curve of the rhizoplast.

When the Leishman-Donovan body is seen as an elongate, oval or torpedo-shaped form, this elongation has usually taken place in an antero-posterior direction, so that the trophonucleus and kinetoplast are left closely approximated in the middle zone of the parasite, the rhizoplast lying in the long axis of the body.

Flagellate Forms.— The orientation of the flagellate form closely conforms to that described for the Leishman-Donovan body, but the parasite is now greatly elongated in an antero-posterior direction and the distribution of parts, as described for the Leishman-Donovan body, is modified in the following respects. The parabasal with the rhizoplast and flagellar vacuole are carried to the anterior extremity of the body and separated from the trophonucleus, but they still maintain an orientation very much as described in the Leishman-Donovan body, i.e., the base of the flagellum (rhizoplast) is near, if not on, the surface, whilst the flagellar vacuole lies deeper in the sagittal plane. The parabasal, if elongated or double, has its long diameter in the transverse horizontal axis. The position of the trophonucleus is difficult to determine. In sections it is most frequently markedly eccentric; the section of the parasite being circular, the trophonucleus, also circular in cross-section, is applied at one point to the periphery of the parasite. In films it is possible to find examples where the trophonucleus appears to be so applied, but on the other hand, even where the kinetoplast is seen at the side, the trophonucleus appears more or less central in position.

SHAPE OF THE PARASITE.

The shape of the flagellate form of the parasite varies considerably, but is on the whole fusiform with the anterior end, from which the flagellum arises, more bulky and obtuse than the posterior. Many developing forms are subglobular. Division forms are often spindle-shaped. Certain forms are almost spirochæte-like, the body being very little broader than the flagellum.

Viewed under a high-power binocular microscope, the body of the parasite appears thick and voluminous, like a bladder filled with fluid, as it does also under dark-ground illumination. Folded-over forms that suggest a flattened shape are most probably due to torsion constricting the parasite.

Not uncommonly the body exhibits a twist in its length, giving a corkscrew-like effect.

THE CYTOPLASM.

The cytoplasm, as seen in wet fixed preparations stained with iron hæmatoxylin, is finely vacuolar or reticular in structure, and a few small, more distinctly visible vacuoles are generally present, especially posteriorly. In certain forms, considered degenerative, vacuolization is distinct. In Giemsa preparations most parasites contain a variable number of granules which take on a stain with Giemsa very similar to chromatin (azurophil). These granules are most numerous in the posterior portion of the body, but also occur anteriorly. The granules are also numerous and conspicuous in the round forms.

Almost invariably in suitably stained preparations there is present in the fully-developed flagellate in the anterior portion of the body an obliquely placed clear cleft-like streak. This passes from the neighbourhood of the trophonucleus obliquely forwards towards the flagellar vacuole. Somewhat similar streaks may sometimes be seen in other parts. The appearance and

situation are certainly suggestive that this cleft may be of the nature of a cytostome.

A peculiar appearance often shown by the cytoplasm is a longitudinal division of the body of the parasite into two portions, one more lightly staining than the other, or in Giemsa preparations showing a different tint. The appearance is due to different structures forming the body (*vide* axostylar body).

In addition to the recognized organellæ, there are frequently to be seen in the cytoplasm of both the fully-developed flagellate and of the stumpy forms certain rather obscure lines, chromatinic in character.

Two different lines appear to be distinguishable in the anterior portion of the body. One of these is a line passing more or less directly from the neighbourhood of the trophonucleus to the neighbourhood of the parabasal. The other is a line passing from the neighbourhood of the trophonucleus apparently to join the flagellum. A dark line may also often be seen extending posterior to the nucleus. The lines appear to be due to structures about the base of the flagellum connected with the axostylar body or its rudiment.

THE ORGANELLÆ.

The Trophonucleus. The trophonucleus is typically circular or oval in shape. In elongate forms it may be compressed so far as to appear linear. With a moderate degree of differentiation of the iron hæmatoxylin preparation the trophonucleus appears as a clearly differentiated circular or oval area. In some cases this may be surrounded by a paler zone of cytoplasm. With further differentiation the black area, which represents the whole nucleus, becomes resolved into a more or less central dark portion, karyosome, and a lighter staining peripheral portion. The trophonucleus varies in appearance. Two types of nuclei can be distinguished, depending upon the condition of the karyosome. In Type I nucleus, the karyosome is present as a conspicuous globular body approximating to one-half or one-third of the diameter of the nucleus. In Type II nucleus, the karyosome is more delicate in appearance, as well as less simple in form. In this type of nucleus the karyosomic mass is associated with a rod-shaped structure (centrodesmose). The appearances for the most part can be placed under one or other of the following heads : (*a*) The rod is unilaterally situated in respect to the karyosomic mass, giving a drumstick effect. (*b*) There is a rod with a medianly situated karyosomic mass (Saturn-ring effect). (*c*) The karyosomic mass is divided and the two portions lie at either end of a centrodesmose (dumb-bell effect). (*d*) Various intermediate effects, e.g., a short unilateral centrodesmose.

The two types of nucleus are commonly bridged by intermediate appearances. Thus, in a nucleus otherwise of Type I character there may be a minute accessory granule, or there may be two such granules. A very common appearance is such a granule joined to the karyosome by a short connecting rod. In addition to the above there is commonly seen a large karyosome mass associated with a smaller mass of perhaps half its size, or there are two

closely approximated nearly equal-sized masses, or a bilobed effect of the karyosome. Occasionally the karyosome has the appearance of a cross.

All the appearances are the result of different arrangements of the following: (*a*) A nuclear membrane enclosing nuclear material; (*b*) a karyosome mass which undergoes division; (*c*) an intranuclear centriole which may be present as a single body, or as two bodies when divided, one or both of which may be external to the karyosome; and (*d*) a centrodesmose joining the two centrioles when present.

Kinetoplast.—Includes all the structures connected with the base of the flagellum: (*a*) A clear vacuolic area, flagellar vacuole; (*b*) the parabasal; and (*c*) the base of the flagellum, rhizoplast, with, under certain circumstances, a terminally situated granule; (*d*) blepharoplast.

Flagellar Vacuole.—At the base of the flagellum is a clear vacuole to be seen in the living condition and in stained preparations, *flagellar vacuole.* The flagellar vacuole is generally about half the width of the parasite in diameter. Most frequently it is seen occupying a position at one side of the body, in which case the extreme basal portion of the flagellum passes around and external to it. It is extremely conspicuous in the early stages of development of the flagellate. On the nuclear hypothesis, the flagellar vacuole would represent the actual nuclear sac.

Parabasal.—The parabasal is a body about half the diameter of the trophonucleus. It is usually in the form of the two equal-sized, closely approximated or connected masses, but is sometimes rod-shaped or may appear as a globular, triangular or heart-shaped mass. The parabasal appears to be very closely connected with the flagellar vacuole, and may be described as lying on the dorsal wall of this structure, usually occupying a posterior position.

Flagellum.—The flagellum is seen as a more or less uniformly staining untapered filament. Its apparent width varies with fixation and staining. At its base the flagellum passes round the flagellar vacuole on its dorsal aspect, and usually appears to join one side of the parabasal. As a rule, especially in wet fixed films, the flagellum appears to pass into the parabasal without any break. In certain cases, a minute gap may be seen between the abrupt termination of the flagellum and the parabasal, the appearance being as if a small vacuole were interpolated in the substance of the flagellum at its extreme base. Under certain circumstances a minute stained particle can be seen at the basal termination of the flagellum, *blepharoplast.* Within the cytoplasm the flagellum becomes rather suddenly somewhat thinner, where it has the appearance of passing along the wall of the flagellar vacuole. Where the sheath of periplast is continued on to the flagellum, it often stains rather darkly, giving the effect of a funnel-shaped collar round the base of the flagellum. In its passage over the flagellar vacuole the extreme base of the flagellum, instead of being constricted, as just described, is often considerably expanded, so that it appears to end in a gradually broadening cone in its passage towards the parabasal. This appearance is associated with division. At its free terminal extremity the flagellum often shows a slight club-shaped thickening very suggestive of an organ of attachment.

Axostylar Body of Christophers, Shortt and Barraud.—A body that can scarcely be given any other designation exists in this parasite. Especially in Giemsa preparations, the body of the parasite frequently shows differentiation of the cytoplasm in such a fashion that a distinct ribbon-like darker stained band can be seen passing from the parabasal dorsally over the trophonucleus to the posterior portion of the parasite. In most preparations where there are many parasites, a number of peculiar bodies may be seen which consist of flagellum, a parabasal granule and a sharply defined fusiform prolongation. This prolongation is usually much the same shape as the body of a parasite, but of about half the thickness. The whole structure in question may be mistaken for a flagellate. It will be observed, however, that the fusiform structure resembling the body of a parasite has no nucleus. It is a surface structure rather than a core.

As a result of study of the apparent time relations between various details of division of the parasite during culture, the following schema has been constructed by Christophers, Shortt and Barraud, it being understood that in the schema the two newly-formed parasites after stage 11 are again at stage 1 :—

SCHEMA OF A CYCLE OF DIVISION CHANGES.

Stages	Parabasal	Flagellum	Trophonucleus	Cytoplasmic body
1	Single or bilobed with median rhizoplast	Single	Without change	Fusiform
2	Bilobed with lateral rhizoplast	Ditto	Ditto ...	Ditto
3	Ditto	Changes in rhizoplast	Ditto ...	Ditto
4	Bilobed or divided	Ditto	Early division changes	Ditto
5	Divided	New rhizoplast formed	Rounded-up ...	Pear-shaped
6	Ditto	New flagellum about half length of old	Centrodesmose	Globular
7	Ditto	Ditto	Band-like ...	Ditto
8	Divided and widely separated	New flagellum about three-quarters length of old	Trophonucleus divides	Ditto
9	Each single or bilobed with median rhizoplast	Each single ...	Type I nucleus	Cytoplasmic body divides
10	Each bilobed with lateral rhizoplast	Ditto	Without change	Each pear-shaped
11	Ditto	Each single or changes in rhizoplast	Ditto ...	Each pear-shaped or fusiform.

TYPES OF PARASITES SEEN IN CULTURE.

I.—Unaltered Leishman-Donovan bodies. These are found in early culture periods only.

II.—Early flagellation forms. These may show all stages of flagellar formation short of the full development of the flagellum. These forms are globular or ovoid in shape. They vary in size from those little larger than a Leishman-Donovan body to large predivision forms.

III.—Stumpy forms. Short subglobular, pear-shaped, oval or very

frequently broad, symmetrical spindle-shaped forms with well-developed flagellum.

IV.—Fusiform multiplicative forms: (*a*) Elongate oval or torpedo-shaped forms, (*b*) spigot-shaped forms; with length exceeding three times the breadth; body with anterior and posterior portions not very dissimilar (*a* form), or with posterior portion more attenuated (*b* form); flagellum as long as body or longer.

V.—Mature flagellates. Large flagellates with length five to six times the breadth.

VI.—Globular or subglobular flagellate forms. Very small forms without a flagellum or in process of losing this and with (usually) one chromatin mass of relatively large size. As a possible resistant or encysted stage, this form has considerable importance.[1]

(7) Degenerative forms. The following types of parasite appear to be degenerative forms: (*a*) Large fusiform or rhomboidal; forms with vacuolated cytoplasm and often a straight flagellum. (*b*) Pyriform bodies, with vacuolated cytoplasm and degenerative appearance with long flagellum. (*c*) Large globular forms, 3 microns or more in diameter, often with irregular nucleus and flagellum several times diameter of the body.

LIFE CYCLE OF THE PARASITE IN CULTURE.

In liquid medium, up to the second day, only developing aflagellate forms (Types I and II) were present. On the third day there appeared in addition to these a number of Type III (stumpy forms) and Type IV (fusiform multiplicative forms). From the fourth day onwards the dominant forms were Types IV, V and III, of which the first two were in general in great excess, Type IV reaching a maximum before Type V. The aflagellate (cystic ?) forms (Type VI) are not seen until the eighth day. As regards the mature flagellate forms (Type V) a few were seen as early as the third day, but this form reached its greatest numerical prevalence rather late in the culture, i.e., on the eighth day.

In the first stages of the development of leishmania cultivated on N N N medium, Franchini observed that small annular bodies were present.

[1] Christophers, Shortt and Barraud have observed that whilst the general life of the culture has reached a maximum by the tenth day, and has apparently become extinct at a later period, there is one form of the parasite that seems especially associated with the later stages. These are the O bodies described by Row. These forms seem to make their first appearance when the culture has reached its zenith, i.e., about the eighth day or later, and they become increasingly numerous as the culture ages up to a point. In their most recent paper (1927) Shortt and others have shown that all the aflagellate forms found in older cultures, and classed together as O bodies, are not necessarily identical in structure, since some may consist of trophonucleus and cytoplasm only, while others may include, in addition, axoneme and parabasal. In the case of *L. donovani* they believe that the O bodies are end-products of degeneration and are not viable. Attempts to make subcultures of *L. donovani* in which all active flagellate forms had disappeared have always, in their experience, been unsuccessful.

They resembled young malarial parasites. They contained one or more chromatin granules. Nuclear masses and flagella did not develop until later. In older cultures (thirty to forty days) flagellate forms were not so numerous. There appeared in their place a circular form, which Franchini considers to be an encapsulated form. In cultures eighty days old, the circular forms, non-flagellate and without a kineto-nucleus, were the only ones present. These have only one chromatin mass.

Di Cristina and Cannata hold that in young and old pure cultures, two different forms are present. One is an elongate form, and the other a pear-shaped non-flagellate body, the latter being considered by them as being possibly sexually different from the former. Marzinowsky and Wenyon have recognized both these forms in cultures of *Leishmania tropica*. Di Cristina and Cannata also call attention to certain forms which they regard as showing evidence of a sexual process. These are spherical forms containing chromatin granules besides the nuclei. They think that the granules indicate a maturation process in the preparation of gametes which conjugate in anisogamy and give rise to aflagellate forms.

Rogers and Nicolle found that 20°—22° C. was the optimum temperature for the development of leishmania, up to the flagellate stage. Franchini found temperatures of 18° and 22°—28° C. favourable for the development of the parasite. Young and fairly old cultures will grow and develop for a few days at a temperature of 30° C. Between 37° and 40° C the flagellate forms and young cultures die. In old cultures a few of the flagellates may survive a temperature of 45° C. for a short time, and can be subcultured (Franchini). Di Cristina has found that young cultures can withstand a temperature of 37° C. for twenty-two hours, whereas at a temperature of 42° C. they die after one hour's exposure.

At blood heat the Leishman-Donovan bodies die out in a day or two. At 27° C. they live for several days and appear to multiply considerably. At 22° C. the parasites multiply rapidly and also increase in size and subsequently develop into a flagellated stage. Below 15° C the development ceases.

Fresh human serum destroys leishmania (Archibald and Cornwall), and according to Cornwall the serum of goats, sheep and guinea-pigs has the same action. On the other hand, the parasites are not destroyed by the serum of rabbits, dogs, cats and hens. This action of serum is lost by heating it for half an hour at 55° C., and is not restored by addition of complement. Olsen did not find the serum of moribund tuberculous subjects, or of experimental animals, or the cerebro-spinal fluid of healthy men had any destructive action upon leishmania, but he found exudates and transudates had this property.

MEDIA FOR THE CULTIVATION OF THE PARASITES.

Sterility is an essential factor for the development of the parasites; the presence of bacteria, especially staphylococci, destroys the parasites of both Indian and infantile kala-azar. Spagnolio found that the parasites of infantile kala-azar grew in the presence of *Micrococcus melitensis*. Water kills the parasites. Moulds and fungi do not interfere with their development. They

quickly degenerate in the tissues after death. They have been cultured by Archibald from the splenic material three hours after death.

Rogers found that normal or slightly hypertonic saline was most favourable to the growth of the parasites, and that the growth was most abundant in a slightly acid medium.

Later on, Novy and MacNeal suggested agar-agar with rabbit's blood. The parasites develop exceedingly well in tubes of this medium. Nicolle found that the parasites grow best in what is known as N N N agar medium.

The following is the formula for the preparation of this medium, as recommended by Shortt: 14 grm. of agar are soaked in distilled water for twenty-four hours, followed by two changes of twelve hours each. Surplus water is decanted off and 6 grm. of sodium chloride added and distilled water poured in to make up the weight to 914 grm. (In 1923 Shortt recommended the addition of 2·5 grm. of glucose to the above.) "The medium is now autoclaved at 15 lb. pressure for twenty minutes, and when required for use is tubed and again autoclaved. After liquefaction in a water bath and cooling to 52 C., rabbit's blood obtained by cardiac puncture is added to each tube to the extent of one-third the volume of agar used. The tubes are mixed by rolling, and sloped until solidified. They are then placed over-night in cold storage or on ice, after which they are capped and incubated for twenty-four hours at 37 C., when they are ready for use."

Row has used defibrinated rabbit's blood laked with 8 to 10 parts of distilled water and then mixed with 2 parts of 1·2 per cent. salt solution kept in a water bath at 56° C. before the laked blood is added to it. He has also used human blood in place of rabbit's blood.

The parasites can also be cultivated in Kligler's medium, which is made as follows: (1) Mix dextrose, 0·1 grm.; nutrient agar, 10 c.c.; normal saline, 90 c.c. (2) 4·5 c.c. of the above are put in separate test tubes. (3) Sterilize in steam sterilizer. (4) To each tube add ½ c.c. sterile rabbit's blood serum.

Various other media that have been tried need not be discussed here.

STAINING OF FLAGELLATES GROWN ON N N N MEDIUM.

The usual procedure is to wash the flagellates in salt solution, to centrifuge, and stain the deposit of flagellates. The following method is a very simple one, and demonstrates clearly the flagellates which are present in the fluid part of the medium, as well as those in the solid parts (Shortt). The details are as follows:—

(1) A loopful of the fluid containing flagellates is placed on a glass slide.

(2) Four loopfuls of citrated physiological saline are added and mixed thoroughly with above.

(3) The liquid is then spread over an area of ¾″ sq.

(4) It is then fixed for fifteen seconds in the vapour of 4 per cent. osmic acid.

(5) The preparation is dried in air as rapidly as possible.

(6) It is then fixed in methyl or absolute alcohol.

(7) It is washed with distilled water, in order to remove any salt.

(8) The preparation is then stained with Giemsa or Romanowsky.

The last stain, employed in three stages, gives especially good results. The following method of Romanowsky staining, described by Shortt in 1918, gives very brilliant results :—

(1) The preparation is stained for two minutes in eosin solution. Polychrome methylene-blue is added and the two stains are mixed and left for another two minutes.

(2) The preparation is then rinsed in distilled water. It is then stained for half a minute in fresh eosin solution. Polychrome methylene-blue is added, mixed and the stain left for one minute.

(3) The preparation is rinsed in distilled water, and stained again as in (2). Finally the preparation is washed in distilled water, until the correct degree of differentiation is obtained, and then dried with filter paper.

STAINING OF TISSUES CONTAINING LEISHMANIA DONOVANI.

The parasites may be stained in tissues by iron hæmatoxylin or Leishman's or Giemsa's stain (Shortt). The following method of fixing fresh tissues, elaborated by Dr. M. N. Dey (Calcutta), gives very good results in showing the Leishman-Donovan bodies when the sections are stained with Leishman's or Giemsa's stain : Small blocks of fresh tissue 2 to 3 mm. thick, are placed straight into acetone or methyl alcohol (or ethyl alcohol, if the others are not available), which is graded in strength from 50 per cent. upwards. Fixation and dehydration are carried out as quickly as possible by taking the blocks through several bottles of the fixative successively with short exposure in each, and the process completed in about half to three-quarters of an hour. After cleaning in xylol, the blocks are embedded in paraffin. The sections made of these blocks are then stained with Leishman's or Giemsa's stain. Dr. C. C. Basu (Calcutta) has obtained good results by fixing the tissues in Zenker and staining with silver solution.

RARE FORMS OF LEISHMANIA DONOVANI. (SEE TYPES OF PARASITES SEEN IN CULTURE DESCRIBED BEFORE.)

(1) Swollen, distorted, vacuolated forms with nucleus, staining with difficulty, or annular forms described by Cornwall and La Frenais.

(2) Coccal, granule-like bodies, described by Archibald and others and found in the spleen and liver of individuals suffering from symptoms like those of kala-azar. In Archibald's case they resembled large cocci, more or less uniform in shape. They were massed together in cells which were devoid of nuclei, and showed no signs of development even after ten days on the N N N medium, but produced kala-azar in monkeys after injection. They probably represent a stage in the life history of the parasites, or some of them may be degenerated forms. They have been described as "aberrant" forms of leishmania.

(3) "Gangues" of Laveran and Mesnil. They consist of masses of cytoplasm containing a varying number of nuclear pairs, and have been variously interpreted as detached portions of the cytoplasm of large mononuclear cells

containing parasites, reproductive phases of the parasites, or degenerative appearances. Yakimoff states that in one and the same preparation he has been able to trace the whole development of these bodies; they commence with one nuclear pair; the nuclei multiply till a multinucleate body is produced, and this segments into corresponding number of nuclei.

(4) Some observers state that sexual forms of the flagellate occur in culture, and that multiplication takes place by conjugation. This is very doubtful.

(5) In old cultures when the nutrient medium of the parasites is exhausted, oval or spherical forms are found, which when stained appear to possess thicker walls, suggestive of the cystic or post-flagellate stage of certain "herpetomonad flagellates" which on subculture may again develop into flagellates.

(6) A "thick-tailed" parasite, with flagellum four or five times as thick as usual, has been found by Cornwall and others in bed bugs. One might think that they were specific of leishmania developing in bed bugs, but Shortt has also seen them in *Herpetomonas ctenocephali*.

(7) Row described "post-flagellate" and "super-post-flagellate" forms in culture, and he believes that they are most infective to mice. Cornwall doubts this, and Shortt looks upon them as degenerative forms. They consist of small rounded bodies resembling the original parasite, and they probably bear some relation to form 5.

(8) A curious, very slender form has been observed in culture by Leishman, probably subsequently giving rise to ultra-microscopic bodies.

(9) Cystic forms have been described in dried cultures, but they are probably artefacts formed in process of drying.

(10) Small rounded forms with very long flagellæ have been described by Shortt.

According to Wenyon the origin of the structures in (2) and (3) is in the cytoplasm of large cells containing *Leishmania*, portions of which have been broken off. They are merely fragmentation bodies.

LEISHMANIA DONOVANI AND LEISHMANIA INFANTUM.

Morphologically there is no distinction between the *Leishmania* (*Leishmania donovani* and *Leishmania infantum*) giving rise to internal leishmaniasis in different parts of the world where the disease occurs, though in the Mediterranean and Caspian areas the disease is associated with a similar disease in dogs. As stated elsewhere there seems no reason to separate the Mediterranean leishmania from those of India.

CHAPTER IV.

INOCULATION EXPERIMENTS AND LESIONS PRODUCED IN EXPERIMENTAL ANIMALS.

Methods of Inoculation. — Infection is most easily produced by intraperitoneal injection of large doses of the emulsion obtained by crushing an infected spleen, liver or bone-marrow in normal saline solution. In the case of larger animals, intrahepatic or intravenous injection may be made. Infection is not so readily produced by subcutaneous injections. As regards the material to be injected, cultures of the virus or emulsions of the above-mentioned tissues of an infected man or some other animals may be used. Infection is more likely to take place with the virus from the affected organs. In the case of culture a large dose may be required for successful inoculation. Attempts at bringing about infection by oral administration of culture of the virus have sometimes been successful. Grey monkeys have been successfully infected by feeding them with emulsion of the Sudan virus (Archibald). Rats have been infected by feeding them with cultures of the Indian virus (Cornwall and La Frenais). Knowles infected a monkey from ingestion of the infective material. Greig and Christophers obtained positive results by the injection of infective material into the lumen of the intestines in a monkey. (It may be stated here, incidentally, that Laveran infected mice by oral ingestion of *L. tropica*.) Recently Christophers, Shortt and Barraud have observed that, with one exception, where a fallacy may have been introduced, the oral administration over a long series of cultures of *Leishmania donovani*, failed to infect tame mice. Adelheim has observed that a healthy mouse was infected by being kept for five months in a jar containing an infected mouse. The infected mice had ulcers in the intestine containing *Leishmania* and it was thought that infection of the originally healthy mouse was due to oral contamination.

Working with *Leishmania donovani*, Row found that positive results, whether local or general, were obtained only when the cultures abounded in small rounded bodies resembling the original parasites or cocci-like bodies, which he described as "super-posttlagellated forms." His observations seem to differ from those of Nicolle, Manceaux, Laveran, Gonder, and more recently of Shortt, who obtained positive results only by using active flagellates.

Many observers hold that the posttlagellated and super-posttlagellated forms described by Row are products of disintegration of the parasites in the culture. Christophers, Shortt and Barraud have, however, recently observed

that primary cultures of *Leishmania donovani* of an age of six days or less are probably incapable of producing infections in tame mice when inoculated intraperitoneally, and that primary cultures of an age of seven days or more are almost invariably capable of causing infections in tame mice under the same conditions. They have also observed that under the most favourable culture conditions in N N N medium the O bodies of Row (? cystic stage) first appear on the seventh day of culture. It is therefore a somewhat significant fact that cultures of the seventh day were the earliest to produce infections in mice.

Still more recently Shortt, Craighead, Khazan Chand and Swaminath have shown that the flagellate forms of *Leishmania donovani* found in cultures are capable of producing infection when inoculated intraperitoneally into white mice without the presence of the O bodies described by Row, and that all the evidence available at present is against the probability of the O body playing any part whatever in producing infections.

DOGS.

Infantile Kala-azar.—Nicolle, Manceaux and Comte were the first to inoculate dogs with the virus of infantile kala-azar. They found that the disease in the dog was generally a mild one, and there might be transitory elevations of temperature lasting several days. Gabbi and Visenteni (Italy), di Cristina and Cannata (Italy), Jemma, di Cristina and Cannata (Italy), Alvares and Pareira da Silva (Lisbon) subsequently made successful experiments with dogs. Di Cristina and Cannata found that young dogs were more receptive of infection than old ones, and that even a heavily infected dog could continue well nourished and without any detectable disturbance of the organs. Gabbi inoculated successfully an Indian dog with the Mediterranean virus. Novy infected dogs with cultures of the virus obtained in Tunis. He obtained similar results in his experiments on dogs which he made in collaboration with McJunkin and Schule. Laveran (Paris) and Laveran and Pettit (Paris) infected dogs with the Tunisian virus. They made a most extensive series of experiments on dogs, and found that the infection might be sometimes of such a mild nature in dogs that parasites could only be detected in the organs by the cultural method. In these mild cases spontaneous recovery may take place. The spleen is not enlarged in such cases. Nicolle and Blaizot infected a dog by intravenous inoculation with a culture. Yakimoff (Turkestan) infected dogs with the local virus. Pulvirenti and Tomaselli (Catania) successfully inoculated the monkey intrahepatically with spleen juice of an infected child.

Indian Kala-azar.—The earliest experiments to infect dogs with the virus of Indian kala-azar proved unsuccessful (Donovan, Mackie and Patton), and this fact was originally advanced as a proof that Indian kala azar was distinct from infantile kala-azar of the Mediterranean. It is now well known that the Indian virus will give rise to infection as often as the Mediterranean virus if sufficiently large quantities of the virus are used. Dogs were

subsequently successfully inoculated by Donovan (Madras) and by Patton (Madras) by means of large doses ranging from 2 c.c. to 4 c.c. of a thick emulsion of infected material from the liver, spleen, or bone-marrow. Wenyon infected a dog in London from a case of kala-azar from India, the virus being afterwards successfully passed through four successive dogs. Laveran infected dogs with a culture of *Leishmania donovani* obtained from Row in India, by intravenous and intrahepatic injections. Mackie, and Knowles with Napier and Das Gupta have also infected dogs with the Indian virus.

MONKEYS.

Infantile Kala-azar.—Nicolle, Manceaux and Comte (Tunis) showed that monkeys could be readily infected with the virus of infantile kala-azar. In these animals the disease may run a mild or a severe course. The symptoms in the monkeys are more evident than in dogs. There may be fever of one or two weeks' duration, alternating with periods of apyrexia of the same length or longer. When the infection becomes severe the fever is more regular and finally gives place to a subnormal temperature. The spleen may be much enlarged. Nicolle and Manceaux have observed an exact reproduction of infantile kala-azar in one of their experimental monkeys. They also noted that the virus in passing through monkeys became more and more attenuated, and they considered that to preserve the strain in the laboratory it would be necessary to keep it in dogs. Laveran and Pettit infected monkeys and found that some of them acquired a slight infection and others a fatal one. They also observed that sometimes, as in the case of dogs, the infection was a latent one, and the parasites could only be detected in the organs by the cultural method. Tyzzer and Walker produced a purely local lesion by subcutaneous inoculation of a monkey with cultures of the Mediterranean virus. Marshall (Sudan) and Archibald (Sudan) infected monkeys with the Sudan virus.

Indian Virus.—Row (Bombay) produced local nodules in monkeys by rubbing the virus from a human kala-azar spleen into the scarified skin of monkeys, and also by injecting subcutaneously 1·5 c.c. to 3 c.c. of an emulsion from its subculture. The emulsion from one of these nodules was inoculated into the skin of another monkey with the production of a local lesion. It was also injected into a monkey intraperitoneally, giving rise to a general infection. Korke performed similar experiments. He found that subcutaneous inoculation sometimes gave rise to a local lesion in the skin, and at other times to a general infection. In some cases general and local infections occurred at the same time. Patton infected monkeys with large doses of an emulsion of a heavily infected spleen of a kala-azar case. Laveran, working with the Indian virus, found that the nature of infection in monkeys with the Indian virus was similar to that produced by the Mediterranean virus. Mackie infected a monkey with splenic juice of a case that died of kala-azar. Shortt (Assam) infected monkeys with the Indian virus from man and other infected monkeys, and in some cases he produced acute infection in the animals.

JACKALS.

Infantile Kala-azar.—Nicolle and Blaizot infected jackals by intraperitoneal inoculation of an emulsion of the organs of an infected dog.

Indian Kala-azar.—Patton successfully inoculated the jackal with emulsion of a heavily infected human spleen.

CATS.

In spite of many attempts with these animals no one has yet succeeded in infecting them. There is a record of one case of a natural infection of a cat observed by the brothers Sergent, Lombard and Quilichini in Algiers.

MICE.

Infantile Kala-azar.—Laveran and Pettit were the first to infect mice with the Mediterranean virus. Yakimoff and Khol-Yakimoff infected mice by injecting cultures intravenously as well as intraperitoneally. Rutelli also infected mice by intravenous injection of the splenic juice from a human case. Laveran found that the infected mice became anæmic, their spleen became much enlarged and their testicles showed signs of degeneration. He observed that the virus became more and more attenuated in passing through mice, and that the percentage of natural recoveries increased with the successive passage of the virus. In certain experimental mice the infection was present even after a year, and in others the characteristic lesions were still present, though parasites could not be found.

Indian Kala-azar.—Row (Bombay) succeeded in infecting mice with the virus obtained in the following ways: (1) From a local cutaneous nodule in an infected monkey, (2) from the spleen of infected monkeys, and (3) cultures of the virus. Laveran also made successful experiments in mice with the Indian virus. Mackie infected white mice after intraperitoneal injection of the spleen juice of a boy who had died of kala-azar. Shortt produced heavy infection in a mouse with the Indian virus.

RATS.

Infantile Kala-azar.—Archibald found them refractory to the Sudan virus.

Indian Kala-azar.—Patton infected a rat with the Indian virus. Cornwall and La Frenais infected a rat by injection of a culture of the virus, and another by oral administration of bread mixed with the culture. Wenyon infected white rats from an infected man.

GUINEA-PIGS.

Infantile Kala-azar.—Laveran and Pettit were the first to show that these animals were susceptible to the virus of infantile kala-azar. They produced a mild infection in them. Franchini produced a fatal general infection in a guinea-pig by intraperitoneal inoculation with a culture very rich in parasites. Archibald found that guinea-pigs were refractory to the Sudan virus.

Indian Kala-azar. Guinea-pigs have not yet been infected with the Indian virus.

RABBITS.

Infantile Kala-azar. Volpino successfully inoculated the cornea of rabbits by scarifying them and depositing upon them material from the spleen of a dog infected with the infantile virus. Rabbits have been found refractory to the Sudan virus (Archibald). Mantovani produced a general infection in a rabbit by intravenous injection of a culture of the infantile virus.

Indian Kala-azar. Rabbits do not appear to have been infected with the Indian virus.

FLYING FOXES.

These have been successfully inoculated by Mackie.

HAMSTERS.

Recent experiments by Smyly, Young and Brown and Meleney have shown that hamsters seem to be very easily susceptible to inoculation with *Leishmania donovani*. The virus did not become attenuated in its successive passage through them. Meleney has shown that the infection progresses steadily in its intensity in these animals, and at the end of fifteen months the tissue of the spleen, liver, intestinal mucosa, bone-marrow and lymphatic glands is replaced by macrophages containing *Leishmania*.

GERBOA AND GERBIL.

Archibald infected them with the Sudan virus.

It may be stated here that while many species of animals have been successfully inoculated by the virus of Indian or Mediterranean kala-azar, the percentage of successful inoculations to those which were unsuccessful has usually been small. This result has probably been due to the fact that experimental animals which showed no clinical manifestations of infection were not further examined by cultural methods. In this connection it may be stated that Shortt has shown that out of thirteen monkeys inoculated intraperitoneally with splenic material, ten, or 77 per cent., became infected. All these infections except two were detected by cultural methods. In certain cases the infected animal may remain in good health, and scanty leishmania are found on spleen or liver puncture and positive results obtained from culture of the liver or splenic juice for some time after infection. Subsequently the liver or splenic juice showed no leishmania in film or culture. The condition has been termed latent leishmaniasis (Knowles). In other cases the animal may show most of the signs of acute or chronic human kala-azar. Recent experiments seem to point out that the hamster, as stated before, is very easily susceptible to inoculation. Dogs and monkeys perhaps come next in order of susceptibility. Mice are more susceptible than rats. Guinea-pigs and rabbits are more difficult to infect. Cats, as stated before, goats, calves, pigs, have not yet been successfully inoculated.

We give here a summary of the inoculation experiments with the viruses of Indian as well as of infantile kala-azar and their results :—

(1) Subcutaneous injection of the Indian virus is followed by a local or a general lesion or both.

(2) Intravenous, intrahepatic or intraperitoneal injection of the Indian virus is followed by a general lesion, but never by a local lesion.

(3) Subcutaneous injection of the Mediterranean virus may be followed by local lesions similar to those produced by the Indian virus (Nicolle and Manceaux, Tyzzer and Walker).

(4) Intraperitoneal or intravenous or intrahepatic injection of the Mediterranean virus is followed by lesions similar to those produced by the Indian virus.

(5) A monkey after recovering from experimental infection with the virus of infantile kala-azar was found subsequently immune to infection with the virus of Indian kala-azar as well as of infantile kala-azar (Laveran). If these observations of Laveran are corroborated in future, then the results may be conclusive in proving that Indian kala-azar is identical with infantile kala-azar, an attack of one conferring immunity against the other.

(6) Dogs are inoculated with difficulty by the virus of Indian kala-azar, but with ease by the virus of infantile kala-azar.

(7) The viruses of Indian and infantile kala-azar are inoculable into dogs, monkeys, white mice, rats and some other rodents.

THE BIOLOGICAL RELATIONSHIP BETWEEN THE DIFFERENT LEISHMANIA.

It will be seen from what we have stated that more or less identical species of animals have been successfully infected by the Indian or the Mediterranean virus, with the production of more or less identical lesions with only slight minor differences. It appears that as our knowledge of the *Leishmania* has increased, the identity of these two viruses has become more and more close. Laveran believed that it was no longer possible to doubt the identity of the two diseases. We cannot, however, lay much stress upon the value of inoculation experiments alone in determining the various species of *Leishmania* found in man, for *Leishmania tropica*, which causes cutaneous lesions in man, may produce general infection in inoculated animals similar to what is observed in the case of the Indian and the Mediterranean virus. The facts (1) that the virus of Indian or Mediterranean kala-azar may give rise to purely cutaneous lesions in animals and occasionally in man, while *Leishmania tropica* may produce generalized infection in experimental animals ; (2) that the organisms are morphologically indistinguishable from each other ; and (3) that they behave similarly in culture have led some observers to hold the view that *Leishmania tropica* is only a variety of *Leishmania donovani*. If this view proves to be correct, then it may be concluded that *Leishmania tropica* has adopted different conditions of life and transmission from those of the parasites of Indian or Mediterranean kala-azar.

On the other hand, the difference between oriental sore, one of the diseases caused by *Leishmania tropica*, and internal leishmaniasis is so great that one

cannot base such a far reaching conclusion, incontestably, upon the results of these experiments. If, however, the parasite of oriental sore be specifically identical with that of kala-azar, it must have been attenuated in its virulence, for kala-azar is as fatal as oriental sore is benign. Nicolle seems to have succeeded in producing a certain degree of immunity to oriental sore and to kala-azar in dogs and monkeys by injecting them intraperitoneally with cultures and infected material containing the virus of the local and of the general disease, though Patton records the case of a patient who contracted kala-azar after recovery from oriental sore. In support of the above view, it has long been recognized in India that in districts where kala-azar is common, oriental sore is rare. Recent work has shown that this is also partially true for the Mediterranean area.

The position becomes still more complicated from my findings (1923) and subsequently those of others in which it was found that patients after having recovered from kala-azar after antimonial treatment showed, after certain periods, skin lesions containing *Leishmania donovani* and at the same time showed complete sterilization of the internal organs. Still more recently, only local skin lesions containing *Leishmania* have been observed in some cases, coming from certain kala-azar infected parts of Bengal, in which there was no previous definite history of kala-azar (Acton, Gupta). If future observations prove that these are cases of infection by means of the virus of kala-azar then the identity of *Leishmania donovani* and *Leishmania tropica* becomes more and more close. All these cases of dermal leishmaniasis including those described by me may be regarded as infection of the skin, observed in individuals in which the internal organs either have a natural immunity to infection with *L. donovani* or have acquired an immunity by a previous attack of kala azar, cured either spontaneously but not recognized by the patient, or cured after treatment with antimony. Or is it that these cases bear the same relation to kala-azar as vaccinia does to variola, i.e., the Leishman-Donovan bodies in passing through one vector lose their virulence, while in passing through another retain it, giving rise either to a local benign skin lesion in one case or virulent kala-azar in another. We hope that when the transmission problem is solved, the identity or otherwise of the *Leishmania* found in internal leishmaniasis and in oriental sore will be solved. At present we may, perhaps, remain contented with the view that Gabbi put forward that the kala-azar of India, the Mediterranean, the Sudan, China and other countries is always the same disease and "there seems no reason to separate the Mediterranean *Leishmania* from that of India" (Wenyon) and that *Leishmania infantum* as the name of a distinct species lapses in favour of *Leishmania donovani*.

"Attempts have been made to differentiate the species of *Leishmania* by serological tests, the use of which for the separation of true species is of very doubtful value. The most precise statements are those of Noguchi (1924). He employed strains of *L. donovani*, *L. infantum*, *L. tropica* and *L. brasiliensis*. Rabbits were inoculated intravenously on four occasions at five to seven-day intervals. The sera from these animals were then used on

cultures to test their agglutinating power. It was found that in dilution of 1 : 10, or even 1 : 100, the serum of the animals inoculated with *L. donovani* agglutinated this organism and *L. infantum*, but not the two others. Similarly, the serum from an animal inoculated with *L. tropica* agglutinated this organism alone, and the same was true of the serum of an animal inoculated with *L. brasiliensis*. From these reactions it appears that serologically the organisms tested fall into three groups, in conformity with the clinical types of disease produced. If the sera were added to the culture media, they were similarly specific in changing the character of the growth of the homologous organisms" (Wenyon, PROTOZOOLOGY, 1926).

CHAPTER V.

TRANSMISSION OF KALA-AZAR — PROBABLE MODES OF INFECTION.

THE mode of transmission of kala-azar is still unsettled. The development of the parasites into a flagellated stage is undoubtedly an important part of its life cycle, but it has still to be proved that this stage is necessary for the natural transmission of the disease from one person to another, or from an intermediate host to man, or that the development of the flagellate stage in certain insects under favourable conditions is a positive sign that they are to be looked upon as the carriers of the disease.

The view that the problem of transmission has yet to be solved, though somewhat at variance with the view that "only experimental transmission from the sandfly would seem to be necessary to prove finally the rôle of the insect in the transmission of kala-azar" is also shared by Wenyon, who, in 1926, in his PROTOZOOLOGY, remarks that "the problem still remains unsolved."

The following modes of infection may be possible :—

(1) *Direct infection from man to man through lesions in the skin, or by the virus being mechanically carried by an insect from the skin of an infected individual.*

The following factors are to be remembered in this connection :—

(1) Leishman-Donovan bodies have been discovered in the papular eruptions and scrapings from the ulcers in the skin in cases of kala-azar (Christophers).

(2) The peripheral blood of kala-azar patients gives positive results in smears in a larger number of cases than was at first imagined, and in almost every case when cultured on N N N medium.

(3) The recent discovery of dermal leishmanoid by the author and others makes it possible that the skin is the channel by which the parasites escape, or at any rate that they may reside in the skin, even when the internal organs have been sterilized against the leishmania.

All these make the chance of skin-to-skin infection a possibility. A direct contact between the healthy and the infected at some unrecognized stage of the disease, when the parasites are present in the skin in largest number, may lead to the infection, if ulcers or abrasions be present in the skin.

Up to now there are no records of any observation to show that direct infection of kala-azar from man to man has ever taken place.

(*B*) *The possibility of transmission by some intermediate hosts.*

(1) Flies and other insects may mechanically disseminate the parasites from skin to skin, by depositing them on ulcers, by biting with their mouth organs, by their faeces contaminating ulcerated or injured surfaces, or by the insects themselves being crushed on the skin (Patton). The insects may also infect food by themselves, or by their excreta, and thereby the virus may gain access to the human body. In these modes of infection it is not necessary to assume that any further developmental changes take place in the insect.

(2) Man may be infected by the natural herpetomonad flagellates in some of the ways indicated under (1). The parasites may pass from insect to insect, from man to insect, and then to man again. The leishmaniform stage in man may indicate that man is not a very favourable medium for the life of the parasite.

(3) Insects may be infected by feeding on the blood, faeces, urine, or other excretions of the infected man, or some vertebrate animal, not hitherto recognized as harbouring the parasites, and then infect a healthy person.

(4) Insects may absorb the virus of the disease by feeding upon the dead body of another host or on its excreta.

The discovery of the development of flagellates of the leptomonas type from *Leishmania* bearing close resemblance to natural insect flagellates has led to the conclusion that they are transmitted to man through a blood-sucking arthropod. Is it possible that more than one blood-sucking insect may transmit the disease, and that the virulence of the virus depends upon the intermediary host through which it passes?

The following gives an account of the experimental work on insects for the determination of the transmitting host.

The first experiments in India in this connection were made by Patton in Madras, in 1907. He used the following insects: *Cimex lectularius* and *rotundatus*, *Pediculus capitis* and *corporis*, *Culex fatigans*, *Anopheles stephensi*, *Ornithodorus savignyi*, *Stegomyia ingens*. Apart from the first two, from the work he published, it appears that he was only able to find the parasite in the alimentary canal of a head louse, but it did not develop there. Experiments with the other insects led to negative results.

Bugs.—In the case of *Cimex lectularius* and *rotundatus*, Patton found that the parasite developed in a manner comparable only to its development in artificial culture media.

When the bugs were allowed to suck the blood from a patient suffering from kala-azar, he was able to find the parasite in the alimentary canal of the bug, in all stages of development up to the flagellate form. He found that on the second day the *Leishmania* increased in size, the two nuclear masses became denser and showed signs of division.

Flagellae were then formed, or the two nuclear masses divided, until a rosette stage was produced. Later on, the single flagellate forms divided by longitudinal fission, producing ovoid and spindle-shaped forms. The ovoid and spindle forms which resulted from longitudinal fission then produced thin irregular-shaped bodies. This occurred on the third day after the blood had been sucked by the bugs. Growth continued until the fifth day, and

after this only flagellate forms were to be seen. Patton thinks it possible that the ovoid and spindle-shaped bodies can be produced by the rosette form, and that the elongated flagellate parasites develop from these.

Donovan repeated these experiments in 1909, but he did not obtain the same results, although he allowed the bugs to feed on patients who had numerous *Leishmania* in their peripheral blood. After ten days he was unable to observe any change in the *Leishmania* ingested by the bugs. He was not satisfied with this result, so he set out to find "a blood-sucking insect which, unlike the bug, is not ubiquitous, but confined in its distribution to regions where kala-azar is prevalent." He thought *Conorhinus rubrofasciatus* was such an insect. But he was not able to effect transmission of the parasite by means of this.

In repeating his experiments in 1910-11, Patton found that when *Leishmania*, from a patient with leishmaniasis, had developed in the alimentary canal of a bug, and the bug was then allowed to suck blood a second time from the same patient or an infected monkey, the flagellate forms, which had already begun to appear, were destroyed.

Patton obtained the development of *Leishmania donovani* up to the flagellate stage in the alimentary canal of *Cimex lectularius*, fed on kala-azar patients, at temperatures which were most favourable for artificial cultures. He studied the changes in the post-flagellate stage and came to the conclusion that the small ovoid bodies which were formed were the infective agents, and probably these were regurgitated from the stomach of the bug in the process of sucking the blood.

Wenyon looked upon these experiments as showing that the stomach of the bug may in a certain measure act as a culture tube for the parasites. They may develop therein, but this does not prove that the bug is the real intermediary host.

Repeated experiments made by the author to find Leishman-Donovan bodies in the stomach of bed-bugs caught in the beds of patients suffering from kala-azar in his wards in the Calcutta Campbell Hospital always failed, though he examined several hundreds of such bugs. Similar negative results were obtained by Franchini. Mackie found that 815 bugs, caught in the bedding of kala-azar patients and 588 laboratory-bred bugs fed on kala-azar patients and injected into monkeys after being crushed gave negative results.

Recently, an entirely negative result was obtained by the Kala-azar Commission, India, on several occasions where bugs were fed on kala-azar cases. The experiments of Shortt and Swaminath, also of Nicolle and Anderson, prove that bed-bugs are not vectors of kala-azar.

Attempts to infect citrated rabbits' blood in test-tubes by causing bugs infected with cultures of *Leishmania donovani* to bite through a membrane consisting of rabbits' skin have given negative results, as shown by culturing the blood on N N N medium in which no flagellates could be developed.

Regarding the period during which infected bed-bugs show *Leishmania* in their stomach or remain infective, the following observations were made :—

Cornwall, and La Frenais showed that the flagellate form of *Leishmania donovani* could live and multiply in the alimentary canal of a bug for at least

twenty-nine days after ingestion of a culture of *Leishmania donovani*, if fed on rabbits' blood.

Patton and Rao fed bugs on regularly arranged cultures of *Leishmania* on N N N media. They found they could subculture *Leishmania* from the middle portion of the gut of one of the bugs at the end of thirty-one days. In the case of another experiment they found they could subculture *Leishmania* from the posterior portion of the gut thirty-four days after the bug had been fed on the culture. When the bugs were allowed to suck blood after they had been fed on culture, it was possible to breed *Leishmania* from the middle portion of the gut, forty-one days later. It therefore seems likely that the parasites can live in the bug for a long time, and one may perhaps assume that bugs are actually the hosts of kala-azar parasites. It must, however, be remembered that according to Patton, La Frenais and Rao, other flagellates can also live in the bodies of bugs for many days, e.g., *Leptomonas pulicis*, *Crithidia ctenocephali* and *Herpetomonas muscarum*. The last mentioned can live in the body of a bug at least forty-five days.

An observation has been made by Mackie, who found that the bugs fed on kala azar spleen juice were infective to mice for seventeen days if the intestinal contents of such bugs were injected into the mice intraperitoneally. Shortt also found that white mice could be infected intraperitoneally by bugs which contained *Leishmania*, nine days after feeding on a kala-azar patient.

Mrs. Adie, from her observations, thought that she found that a Leishman-Donovan body taken in by a bug actually penetrated the walls of its stomach and there developed, much in the same way as rat trypanosomes in the flea. It appears, however, that this process was only observed by her in dead bugs, where obviously the cells behave differently from those in the living organisms. Patton has confirmed Mrs. Adie's work, but it is still not certain that in the living organisms the parasite takes up an intracellular position. Patton thought that the importance of *Cimex* as the invertebrate host of *Leishmania* was proved by these experiments.

Patton's more recent opinion was that the infection in the human subject was not caused by a bite, but by the bug being crushed against the skin.

In his experiments on bugs fed on *Leishmania* cultures, Cornwall was quite unable to recognize any intracellular stage. He thought that the "thick-tailed" stage was merely an abnormality, which had no place in the regular life cycle of the parasite. He also suggested that the time had come to make a diligent search for some carrier of the disease other than the bug.

More recently Mrs. Adie wrote that she found the parasites in the salivary glands and ducts of a specimen of *Cimex rotundatus* taken from the bed of a kala-azar patient. She therefore thought that the disease was transmitted by the bite of a bug. According to Christophers and Shortt these parasites were not *Leishmania* at all, but were organisms belonging to the species *Nosema*. Similar organisms are also found in fleas (Liston, Shortt).

Patton's hypothesis of the transmission of kala-azar by bugs is therefore not yet proven.

Conorhinus rubrofasciatus.—With regard to *Conorhinus rubrofasciatus*, negative results were obtained by Donovan after feeding them on kala-azar cases. Patton found no evidence of development of *Leishmania* in these. Cornwall found the common species refractory, even when fed on cultures. Recent experiments by the Kala-azar Commission, India, have also given negative results.

The facts regarding the bug theory may be summarized as follows: In favour of the theory are:—

(*a*) The developmental stages of the parasite in bed-bugs.

(*b*) The peculiar sporadic nature of the disease tending to be limited to houses.

(*c*) Greater prevalence of the disease in insanitary localities where bugs are likely to be more common.

The following are the objections against this theory:—

(*a*) The developmental forms may be observed in test-tubes under favourable conditions. Beyond the developmental forms of the parasite being present in bed-bugs, there is no further experimental evidence that bugs are carriers of the disease.

(*b*) It is doubtful whether bugs are more common in houses of rural areas in Bengal and Assam than those of towns, but the disease is more common in the former places.

(*c*) All houses in an endemic area are not affected, though bugs may be equally prevalent in all of them.

(*d*) There is no known cause for bugs to become more infective from time to time, and thus to give rise to epidemics.

(*e*) No case of infection has been definitely established in hospitals where kala-azar cases are treated extensively, though bugs are present in them.

(*f*) The geographical limits of the prevalence of the bed-bug are much greater than those of kala-azar in the provinces of India.

(*g*) Kala-azar patients may constantly come from endemic areas of India into other districts where the disease is unknown. Despite the great frequency of bed-bugs the disease has gained no foothold there.

Recently, Shortt, Barraud and Craighead have observed that *Conorhinus rubrofasciatus* in various stages of development, fed on cases of kala-azar, yielded on search no developmental forms of *Leishmania donovani*.

Lice.—Experiments with lice (*P. capitis* and *P. corporis*) have given only negative results. Mackie examined 1,172 body lice and 1,170 head lice taken from kala-azar patients with negative results. 856 crushed body lice and 1,130 head lice from the same source were also injected subcutaneously into experimental animals with negative results.

Fleas.—When Nicolle discovered canine leishmaniasis shortly after human leishmaniasis had been recognized, he assumed that the latter originated from the former. He maintained that the *Leishmania* found in human beings and in dogs were identical, although he had no certain proof that they

were. The experiments by which he tried to transmit the disease from one dog to another by means of ticks, fleas and mosquitoes were unsuccessful.

Basile made several investigations to determine the relationship of dogs to human leishmaniasis. He conducted his investigations on the following lines: (1) Attempts at infecting fleas with human *Leishmania* and healthy dogs by means of infected fleas; (2) study of the *Leishmania* in fleas. In 1911 he announced that he found that dog fleas fed upon splenic juice of kala-azar patients developed cultural forms of the parasite.

In the gut of fleas taken from infected dogs organisms were found which he thought were *Leishmania*, because they showed flagellate and non-flagellate forms resembling those of *Leishmania donovani*. However, it did not occur to him that the ordinary parasites, *Leptomonas* of *Pulex serraticeps* (*ctenocephali*) and *irritans*, were indistinguishable from flagellate and non-flagellate *Leishmania*. He further made the following experiments: (*a*) He kept dogs, found to be free from *Leishmania* by examination of the bone-marrow, shut up with infected dogs and waited to see if the former became infected. (*b*) He took fleas from infected dogs and allowed them to bite young dogs, whose bone-marrow had previously been examined microscopically and found to be free from *Leishmania*. The young dogs became infected. He thought that by these experiments he had established his hypothesis that fleas were the carriers of the disease.

In the first series of experiments he obtained positive results in nearly every case. Almost every dog became infected. However, in similar experiments performed by Massaglia, Marshall, Wenyon, Pereira da Silva and Gabbi, negative results were always obtained. Their experiments, especially those of Wenyon and Pereira, were carried out with greater accuracy. In addition to making microscopical examinations of the bone-marrow, they made certain that no parasites were present by examining cultures made from it. Basile, in his experiments during 1910-11, neglected to do this.

In the second series of experiments, Basile was also successful, but Wenyon, Marshall and Pereira da Silva, who repeated the experiments under more careful conditions, obtained negative results. Sergent, Lhéritier and Lemaire were successful in only one experiment of this kind. They took a bitch who was considered by them to be free from *Leishmania* on microscopical examination of her liver tissue, and transferred eighty-two fleas from an infected dog to her. The animal was subsequently found to be infected. The fact, however, that they assumed that there was absence of *Leishmania* in their experimental bitch before the fleas were fed on her, simply on the evidence furnished by microscopic examination of the liver juice, throws doubt on the accuracy of the result which they obtained, for absence of *Leishmania* can only be determined with certainty when a culture is made. In some cases where dogs are actually infected, no parasites are found in the liver in smears. Much importance cannot, therefore, be attached to the results of this experiment.

In later experiments Basile claimed to have found flagellate forms of *Leishmania* in the human flea. Besides, he took fleas from infected children

and dogs, and injected their intestinal contents into dogs which had been regarded as healthy on the evidence provided by microscopical examination of their bone-marrow. He thought that he obtained positive results.

Pereira da Silva, however, repeated these experiments and obtained negative results, though he had been able to recognize organisms with flagellæ resembling *Leishmania* in the gut of the fleas which he used. His results are summarized as follows :—

(1) He found organisms like *Leishmania* in fleas when they had been allowed to feed on healthy dogs erroneously thought to be infected.

(2) The intestinal contents of a *Pulex irritans* taken from a child suffering from kala-azar, and containing flagellate parasites, were introduced into a healthy dog without producing infection in the dog. He had undoubted proof (by means of cultural examination of the bone-marrow) that his experimental dogs were originally healthy.

(3) Twenty-five fleas (*Pulex irritans*) which were shut up for forty-five days with a dog suffering from severe leishmaniasis did not become infected.

Basile thought that the following experiments provided further support for his hypothesis. He took the intestinal contents from fleas which had been feeding on infected dogs, and which contained parasites of the same type as *Leishmania*, and injected them into the abdominal cavity of the mice. He obtained positive results. Laveran and Franchini, however, using intestinal contents from fleas containing natural herpetomonads, were also able to infect mice in the same way. They found non-flagellate parasites resembling *Leishmania* in the tissue fluids of the mouse. They also succeeded in infecting dogs with the natural herpetomonads of insects.

Basile still contends that the flea is the transmitter of kala-azar, but it can do so only when the requisite conditions of temperature and also perhaps atmospheric humidity exist. He says that the authors that have met with negative results did not pay attention to these during their experiments.

It is well to remember that Basile himself produced no evidence that the flagellates observed by him in the fleas were other than natural flagellates of these insects. The negative results of other observers may have been in large part due to their excluding such natural flea infections. Moreover, the percentage of leishmania-infected fleas discovered by Basile corresponds very closely with the percentage of naturally infected fleas in kala-azar-free countries, so that the inference is that the flea flagellates supposed by him to have developed from the *Leishmania* were really nothing more than the natural flagellates of the fleas.

The following additional facts stand in opposition to the theory that the flea is the transmitting agent of infantile kala-azar :—

(1) *Pulex irritans* and *serraticeps* do not become infected when fed on cultures of *Leishmania*.

(2) The intestinal flora of the flea is an unfavourable medium for the development of *Leishmania* (Scordo). The ordinary intestinal bacteria inhibit the development of *Leishmania donovani* in pure culture (Spagnolio).

(3) The parasite has been found in the peripheral blood-stream of man.

If, therefore, the fleas common to man are the carriers of the disease, one would expect it to be very much more widespread than it is, both among men and dogs.

(4) Infected dogs are hardly ever found in houses where children have contracted kala-azar (Gabbi, Di Cristina, Pulvirenti, Spagnolio and others). In fact, some of these families do not keep a dog at all.

(5) Dogs infected with leishmaniasis are sometimes found in districts and towns where kala-azar has never been observed.

(6) Cases of kala-azar are often observed in households where the dogs are not infected with leishmaniasis.

(7) In certain towns and districts, few children and many dogs contract the disease (Tunis and Algiers). In Catania, Messina, Palermo, Athens, and Pyraeus this proportion is reversed.

(8) In one family a number of children may become infected, but a space of a year or more may intervene between the occurrence of each new case. The new cases do not occur until the old ones have quite recovered.

(9) If the disease was transmitted by fleas infantile kala-azar would occur most frequently in summer and autumn, when the fleas are common and active. The fact is, that it occurs most frequently in winter and spring when fleas are rare and hardly bite at all.

These facts suffice to condemn the theory of the flea as carrier of the disease. It must also be added that so far no clear proof has been put forward that the *Leishmania* found in dogs are the same as those found in human beings. The experiments of Scordo, Moldovan and Giugni show that it is not only in their biological reactions that the two parasites differ, but when grown in the same nutritive medium the human and canine viruses develop in different ways. (Wenyon, however, holds that in the present stage it is better to consider the two parasites as identical.)

Mosquitoes.—Franchini found that *Anopheles* of the *claviger* variety would feed on cultures of *Leishmania* and retain them in the gut for several days. From this he concluded that this insect might be a carrier of the disease.

He claims that there must be some condition of the gut of *Anopheles maculipennis* and Culicidæ, including *Stegomyia fasciata*, which favours the development of *Leishmania*.

He performed a number of experiments in Catania. He allowed the anopheles to feed on the spleen juice of children suffering from leishmaniasis, and was able to find numerous parasites in the gut eight days later. These resembled *Leishmania* in degenerate and rosette forms. Most of those which he found were non-flagellate, but he found flagellate forms in one mosquito after four days, whereas he never observed these in mosquitoes which had not fed on spleen juice. He tried to show that the development of these bodies had much in common with that of *Leishmania*, although in some stages the similarity was not always easy to perceive. Gabbi, however, has shown that Franchini's assumptions are quite unjustifiable, because the infantile form of the disease is most prevalent in January, February and March, when very few mosquitoes are in existence.

Scordo, on Gabbi's recommendation, tried to test Franchini's results by very careful experiments. He observed that *Leishmania* found in spleen juice never developed flagellates in the stomach of mosquitoes. This was his crucial experiment. Besides, Scordo found rosette bodies, which Franchini called *Leishmania*, in mosquitoes which had never fed on infected spleen juice.

Mackie examined 266 *Culex* sp. and 18 *Anopheles* fed on patients with negative results.

In the author's opinion further investigations in this connection are desirable.

House-flies.—Up to the present time there is no evidence to incriminate these as carriers. *Leishmania* are quickly destroyed in them. They have, however, been found by Laveran to carry *Leishmania tropica* mechanically with their wings and legs.

Other Possible Vectors.—Ticks have given negative results (Donovan). The Kala-azar Commission, India, examined a number of *Sarcoptes* with negative results. Ankylostomes, *Strongyloides*, leeches, have all given negative results.

OTHER CHANNELS.

(A) *Infection through the Intestinal Tract.*

The peculiar endemicity of the disease in the Sudan and its limited epidemic nature as first established by Archibald, make it possible that the disease may be a water-borne one, the water being contaminated with cysts from infected arthropods. There is the additional fact that the disease, as observed by Archibald, exists more commonly in villages situated near water, than in those further away.

Archibald and others have further succeeded in infecting monkeys by feeding them with emulsions of virus.

Greig and Christophers record a case in which a positive result was obtained "by the injection of infective matter into the lumen of the intestine in a monkey, but the result is not conclusive as, in spite of all precautions taken, infection by the needle track cannot be absolutely excluded."

The fact that in the early typhoidal stage of the disease there is a positive Widal reaction in many cases, as in typhoid or paratyphoid, suggests to the author that the disease may begin as a mixed infection in the intestinal tract. The presence of *Leishmania* in intestinal ulcers, and the recent confirmation of this by Perry, prove that the parasites may be voided in the stools, and directly contaminate drinking water. The parasites may thus be taken up by water. It is, however, only very rarely that the fæces contain Leishman-Donovan bodies. Mackie found some suspicious bodies in a smear of the fæces, and Critien also found the presence of Leishman-Donovan bodies in the mucous stools of a case suffering from kala-azar. Generally speaking, the examination of the fæces gives only negative results. Further research should be carried out in this direction, in view of the fact that Shortt has recently found *Leishmania* in the urine of kala-azar patients, which makes it possible

that the urine may also contaminate drinking water. It must be remembered that the parasite quickly dies in soil or water.

Recently Christophers, Shortt and Barraud have observed that the oral administration over a long series of cultures of *Leishmania donovani* failed to infect tame mice, with one exception, where a fallacy may have been introduced.

(B) *Infection through Intestinal Worms.*

Hitherto there appears to be no evidence for this possibility. No developmental forms of the Leishman-Donovan body have ever been discovered, either in the embryo of *Ankylostoma duodenale* or the adult worm. Animals injected with the embryos have given negative results. Whether their ovaries may be infected by an undetermined phase of the parasite should be investigated, as well as the behaviour of other parasites in this respect.

(C) *Infection from Contaminated Soil.*

The soil may be infected with the stools, urine, or other excretions containing *Leishmania* from an infected individual, and subsequently infect a healthy individual, through an abraded portion of the skin, like infection in the case of ankylostomiasis. But this method of infection is very unlikely, as *Leishmania* die quickly in the soil.

(D) *Infection by Inhalation.*

The chances of infection by inhalation of substances containing *Leishmania* must be very unlikely.

It is possible, as Archibald suggests, that some insects may harbour an undiscovered phase of the parasite which may infect their ovaries and bring about a hereditary transmission, or that it may be found *ex corpore* in some cold-blooded vertebrate host.

McCombie Young has suggested that perhaps the solution of the problem of transmission may be found in an explanation which combines gross fæcal pollution of the site and a life-history of the parasite in some coprophagic or coprozoic vector, or at any rate in the conditions which attend life in an unhygienic environment.

CHAPTER VI.

TRANSMISSION OF KALA-AZAR—*continued.*

THE RÔLE OF THE SANDFLY IN TRANSMISSION.

KNOWLES, NAPIER AND SMITH have observed that females of *Phlebotomus argentipes* fed upon the blood of kala-azar patients showed herpetomonad forms in their gut. Extensive researches have been carried on in this direction by the Kala-azar Commission, India, and the following is a summary of the latest findings of Shortt, Barraud and Craighead of that Commission regarding the development of *Leishmania donovani* in sandflies.

(A) The Interval from the Initial Feed up to the Second Feed.

If a sandfly (*Phlebotomus argentipes*) is fed on the peripheral blood of a kala-azar patient and is kept at a temperature of 28° C., the following series of changes are observed :—

Any variation of more than 1° C. or 2 C. above or below this temperature acts adversely on the length of the life of the fly.

If the fly be dissected at the end of the first twenty-four hours, a few herpetomonad parasites, singly or in small groups, may be found.

At the end of forty-eight hours after the initial feed, some flagellate forms are present, but these are as a rule not fully developed. The forms seen are in many cases sluggishly motile, showing a swaying motion with little attempt at translation. Division forms are common, if not actually the predominating form.

In dissections, made seventy-two-hours after the initial feed, a new phase of development commences, characterized by the presence, for the first time, of elongated free-swimming forms of flagellate. Besides, there is an increase of all the flagellated forms previously present. Multiplication is evidently very active. For the first time there may be seen the initiation of rosettes. The free-swimming elongate forms appear to be endowed with an intense vital activity, capable of carrying them into any situation penetrable by bodies of their bulk. There is at this stage usually a general diffusion of the parasites throughout the midgut. By the end of the third day there are present in the gut of the fly, although in comparatively small numbers, most of the stages of the flagellate which are seen up to the eighth or ninth day.

The stage arrived at by the fourth or fifth day after the initial feed of the fly is one of intensely active multiplication, until the gut becomes a seething mass of active flagellates with a very distinct bias towards the concentration of the flagellates in the more anterior parts of the midgut.

At the fifth day, before a second feed has been taken, a very large number

of flagellates may be found in any situation in the midgut; besides, there is almost invariably a well-marked tendency for the accumulation of the bulk of the flagellates in the more anterior parts of the midgut, and especially in the narrow portion extending backwards through the thorax from the posterior margin of the proventricular fold. Another situation in which a considerable accumulation may take place is posteriorly near the region where the Malpighian tubules open into the alimentary canal. They sometimes ascend the Malpighian tubules and the diverticulum, and comparatively large numbers, but never masses, may sometimes be found in the intestine as far as the anal opening.

The greatest development of flagellates is at or near the extreme anterior end of the midgut.

The flagellates in this situation are arranged in palisade formation, often many layers deep, around the entire circumference of the proventricular fold. The growth in this situation may form a plug, blocking up most of the lumen of the gut. The flagellates composing the plug show a general antero-posterior orientation. In an unusually intense infection it is sometimes found that the flagellates have penetrated through the comparatively constricted œsophagus and have invaded the pharynx, sometimes in considerable numbers. On the fourth or fifth day after the initial feed, the fly will have laid its first batch of eggs and will be ready for a second feed.

(*B*) *The Interval from the Second Feed to the Eighth or Ninth Day.*

If the fly be examined on the day after its second feed, fed on the same case which supplied the first meal, or on an experimental animal, it will be found that once a massive infection of the extreme anterior end of the midgut has been established, there is a progressive extension forwards of the massive growth of flagellates in the posterior extremity of the pharynx.

The diverticulum, which enters the œsophagus, may be invaded by numerous free-swimming flagellates, and at a latter stage there may be a more massive growth at its œsophageal end.

In the earlier stages of the invasion of the pharynx by a massive growth, its posterior extremity is filled up by a dense growth of flagellates, from which free-swimming elongated forms are given off anteriorly, and these show a decided tendency to swim forwards along the gradually narrowing pharyngeal lumen. As time proceeds there is a continuous extension forwards of the massive growth in the pharynx, until the lumen of that organ becomes blocked with a solid plug of more or less stumpy flagellates. Throughout this process there appears to be a continual setting free of elongate forms anteriorly. These tend continually to carry the infection in an anterior direction and may be the infective forms.

When the main mass of flagellates filling the lumen of the pharynx has reached the junction between that organ and the buccal cavity, it apparently finds in the somewhat angular junction of the two cavities a very favourable situation for the formation of an extensive growth.

From the free anterior extremity of this growth numerous elongate free-swimming forms may be seen to break off and proceed anteriorly as

far forwards as the anterior extremity of the buccal cavity, close to the commencement of the biting mouth parts. The stage is arrived at by the seventh or eighth day after the initial infective feed, and the second, third, or fourth after the second feed.

If the preparation from the extruded contents of the lumen of the gut be now examined, it will be found that the flagellates of which it is composed show a very great preponderance of the elongated types.

This type is *par excellence* the wandering type of flagellate. It is the form which gives to the fresh preparation an appearance of seething motility. This is also the predominant type that forms the great bulk of the flagellates of the compound rosette masses often found in the midgut in heavy infections.

In the preparation made from the gut wall itself, the vast majority of the flagellates seen are of the short stumpy form.

Still more recently, Shortt, Barraud and Craighead have noticed massive infection of the buccal cavity in a sandfly, which had received only two feeds, and was examined on the ninth day after the initial feed. The infection extended forwards in an unbroken column throughout the length of the pharyngeal and buccal cavities until it reached the mouth proper, where the biting appendages took origin, the most anteriorly placed flagellates extending even beyond this situation to a position distal to the salivary pump. It consisted chiefly of elongated flagellates, especially in its most anterior parts. It appears that the intensity of infections, and the extent of their development, in any case depend, other factors being uniform, on the initial number of parasites ingested from the peripheral blood, as well as on the time which elapses since the initial feed.

The same observers have also found that the O bodies of Row might be produced in the midgut of the sandfly, after a second or subsequent feed.

They consider that the course of events in artificial cultures of *Herpetomonas donovani* in N N N medium is an accurate recapitulation of the natural course of development in the sandfly *Phlebotomus argentipes*.

Normally, the adult female sandfly takes its first blood meal on the second day after pupation, copulation takes place immediately and oviposition on the fifth or sixth day, the fly dying immediately afterwards, and the larvæ usually feeding on the dead body of the mother subsequently. The ova require a very high degree of humidity, almost saturation in fact, but the pupæ do not; the imago, however, again requires a high degree of humidity.

The fact that the greater majority of fed female flies die after oviposition on the fifth or sixth day would seem to militate against the insect being the true vector of kala-azar; but it has been shown by Shortt, Barraud and Craighead that the main factors in inducing oviposition with continued life of the fly, in *Phlebotomus argentipes* bred in captivity, are a temperature of 28° C., with upward and downward variations of not more than 1° C. to 2° C., and an accurate adjustment of the degree of humidity. Under these conditions a considerable number of fed female flies survive after oviposition, and feed a second and even a third time.

We have given above a summary of the most recent work of the life history of the parasite of kala-azar in the sandfly, by Christophers, Shortt and Barraud.

A short comparison of the infection of *Phlebotomus argentipes* by *Leishmania donovani*, *Ctenocephalus canis* by *Herpetomonas ctenocephali* and of the tsetse-fly by *Trypanosoma gambiense* is given below :—

Infected insect	Direction of infection in the alimentary canal	Infective forms
Sandfly infected by *L. donovani*	*Forwards* from the midgut till it reaches the mouth proper	No infective cystic forms have yet been discovered
Ctenocephalus canis infected by *Herpetomonas ctenocephali*	*Backwards* till the rectum is intensely infected	Cystic (?) infective forms are probably eaten up by larvæ which thereby become infected
Tsetse fly infected by *Trypanosoma gambiense*	The posterior portion of the midgut is infected and subsequently the salivary glands are infected	No cystic i

The problem of transmission of kala-azar by means of sandfly is still in an unsettled stage. The following points may be noted in this connection :—

(1) There is as yet no evidence of hereditary transmission of the infection from one sandfly generation to the next by direct infection of the sandfly ovum *in situ*.

(2) There is no evidence of infection of the larvæ by their feeding on the bodies of their mothers.

(3) Regarding the possibility of the infection taking place through bites of infected sandflies, the latest report of Shortt, Barraud and Craighead (1927) keeps the position unsettled, as seen from their following remarks :—

"Sixty experimental animals, subjected to 184 bites from a minimum of 152 *Phlebotomus argentipes* known to be infected with *Leishmania donovani* have in no case developed kala-azar. The presumption is not that *Phlebotomus argentipes* cannot transmit the disease by its bite, but that the experimental animals used were not sufficiently susceptible to *Leishmania donovani* to become infected, except as the result of a more intensive exposure to infection than that to which they were subjected in the present experiments.

No single human volunteer has so far been infected by the bites of even heavily infected sandflies in any of Shortt's experiments.

In October, 1926, Shortt, Barraud and Craighead gave an account of the occurrence in nature of a *Phlebotomus argentipes* infected with a flagellate morphologically identical with *Leishmania donovani*. The fly was caught in a house at Bakktiarpur, near the Pusa Estate in Bihar.

Recently it has been observed that very rarely *Leishmania donovani* may undergo development into flagellates in *Phlebotomus papatasii*, and it has been shown by the workers in China that they could develop in the mid-gut of *Phlebotomus major*. (See Addendum.)

The fact that *Phlebotomus argentipes* has been discovered in the city of Bombay (McCombie Young, 1927), the meteorological conditions of which city seem to approximate to those of many kala-azar infected places, appears to go against the theory of the possibility of the transmission of kala-azar by this sandfly, as no undoubted case of kala-azar has been reported from Bombay.

CHAPTER VII.

CANINE LEISHMANIASIS—ITS RELATION TO HUMAN LEISHMANIASIS.

NICOLLE tried to find the intermediate host of the virus which attacks human beings. He turned his attention to domestic animals found in towns, chiefly cats and dogs, because he thought that it was in towns that the disease occurred most frequently.

He was struck by the fact that children suffering from kala-azar had often come in contact with dogs. He examined some of the dogs which he thought were affected, and found parasites of a similar form to those found in children affected with kala-azar, in the spleen, liver, and bone-marrow. He therefore assumed that leishmaniasis in the human subject originated from canine leishmaniasis.

In order to substantiate this conclusion, he decided to examine the dogs which had been killed in the slaughter house in Tunis. He found (March, April, May, 1908) four dogs affected with leishmaniasis, among the 222 dogs which he examined. Nina and W. L. Yakimoff examined 209 dogs in Tunis from January 1 to May 23, 1911, and found five affected with leishmaniasis. Nicolle's discovery led the brothers Sergent to follow out canine leishmaniasis in Algiers. From July 15 to October 1 they examined 125 dogs and found *Leishmania* in nine of them. There was a difference in the percentage of affected dogs in Tunis (1·8 per cent.) and Algiers (7·2 per cent.), although in Algiers only a few cases of kala-azar occurred. According to Nicolle this difference was due to the fact that the investigations were made at different times of the year. Sénevet confirmed this opinion. He examined twenty-three dogs in spring and in summer. In the first group 1·6 per cent. were affected, and in the second 8·8 per cent. Gray, repeating the experiments in Tunis between October 16 and December 5, found only two dogs which were affected.

Basile repeated Nicolle's investigations in Bordonaro, where Gabbi had discovered an endemic area of kala-azar. He found that 71 per cent. of the dogs were affected with leishmaniasis. This, however, was not all. Whereas Nicolle had only been able to confirm the simultaneous outbreak of kala-azar in children and dogs in a few cases, Basile found that in almost every house where a child was affected, a dog also had the disease.

On the other hand, Spagnolio, who had studied all the cases of kala-azar in Bordonaro for four years, did not find, even on a single occasion, a child

and a dog simultaneously affected. Many families in which a child was ill had no dog at all, or if they had one, it was absolutely healthy.

Giugni, using experimental puncture of the liver as a mode of investigation, was not successful in his search for parasites in healthy dogs taken from houses where a child was ill.

Basile found 27 per cent. of the dogs in Rome affected, while human subjects were not affected in proportion. Salvatore made microscopic and cultural examinations of the bone-marrow of ten dogs, and did not find any of them affected with leishmaniasis.

Pulvirenti examined eighty-five dogs in Catania, during the months of February, March, April, 1910. None of them were affected.

In October, November and December of the same year he examined 185 dogs; of these only three had leishmaniasis. Panto in Catania examined 165 dogs; of these only four were affected.

In Malta in April and May, 1910, Critien examined fifty-two dogs and found the parasite in seven. Babington found it in one out of eight dogs examined. Wenyon also found infected dogs in Malta.

In Athens, from December, 1910, till May, 1911, Kardamatis found nineteen out of 284 dogs affected with leishmaniasis, and in Pyraeus fifteen out of 184 dogs had the disease. In another report he collected the observations made on 589 dogs which he examined in the course of the year (December 1910-11). Of these eighty-one had leishmaniasis (13·8 per cent.). The infection reached its greatest intensity during the months of June and July.

Sangiorgi examined 310 dogs in Turin; only one of them appeared to be infected. (Kala-azar has hitherto never been observed in Turin.)

In Hydra Lignos found eight dogs affected out of forty-eight which he examined. The infection was most marked in the summer.

Alvarez and Pereira da Silva examined 300 dogs in Lisbon; nine of them appeared to have the disease. Dschunkowsky and Lubis found one case in the neighbourhood of Elizabethgrad.

Di Cristina and Di Giorgi in Palermo found only two dogs infected out of 1,000 which they examined.

W. Yakimoff and N. J. Schockhor examined 647 dogs in Taschkent and Samarkand in April to September, 1913, and found 157 suffering from leishmaniasis. In this district the total number of cases of kala-azar found amounted to thirty-one, of which twenty-seven were children, and four, adults.

More recently similar observations have been made in Marseilles (Pringault and others), Teheran (Nelgian), West Africa (Lafont and Heckenroth) and Sardinia (Sotgin). No case of kala-azar was ever observed in these places in a human being.

In Hamburg Fülleborn examined fifty dogs; none of them were infected. In Baghdad Wenyon examined 110 dogs and found none of them infected. In Gallebat (Egypt, Sudan) Bousfield found an infection resembling that of leishmaniasis in the spleen of a dog.

In India, Patton, Donovan, Row, Mackie, and many others found that no dogs were infected with *Leishmania*. Recently, however, Avari and Mackie

have found *Leishmania* infection in a dog from Bombay, and another from the Punjab.

In Madras, Donovan examined 1,150 dogs, Donovan and Patton 2,000 dogs without finding any infected. Mackie examined 93 dogs in Nowgong (Assam) and Wenyon 250 dogs in Colombo with similar negative results. Row also got negative results in Bombay, except that quite recently he observed a case of cutaneous leishmaniasis in a dog.

Labbé, Tarcheta and Ameuille, as well as Pringault found definite cases of infantile kala-azar in the South of France. Labbé, Tarcheta and Ameuille, found splenic tumours, and changes in the bone-marrow of the house dog in one of the families where a child had kala-azar. However no parasites were to be found in the dog. Pringault found that canine leishmaniasis occurred in Marseilles.

Relation of Human to Canine Leishmaniasis.—Our knowledge with regard to the above may be summarized as follows:—

In dogs the infection reaches its greatest intensity in the summer, whilst infantile kala-azar of the Mediterranean occurs at the end of winter and in the spring (Gabbi). Besides, there is no obvious parallelism between human and canine leishmaniasis. This is especially the case with Indian kala-azar, as in India very rarely leishmaniasis has been observed in dogs, even in the most heavily-infected regions. Thus, Mackie found that the microscopical examination of the spleen and bone-marrow of over one hundred dogs selected from kala-azar villages in Assam did not reveal the presence of *Leishmania* in any of them. In the author's experience there is no connection whatever between dogs and kala-azar as it occurs sporadically in different parts of Bengal, because very rarely does one find any association between dogs and infected individuals. Even in the case of Mediterranean kala-azar, though in the region where infantile kala-azar occurs, the natural infection of dogs is not uncommon, yet there is great irregularity in the distribution of canine leishmaniasis and infantile kala-azar, and it is only rarely that dogs suffer in houses in which human beings are infected.

On the other hand, dogs may be infected with infantile and Indian kala-azar as well as with canine leishmaniasis by experimental inoculation. They may also be infected by subinoculation. Dogs infected with experimental canine leishmaniasis display close resemblance as regards symptoms and pathological lesions to the disease brought about by experimental inoculation of dogs by *Leishmania donovani* (Laveran). These experiments tend to show that canine and human leishmaniasis may be due to one and the same parasite.

"The important question arises as to whether the naturally occurring disease of dogs is due to *Leishmania donovani* or to some other species. The disease produced in dogs by inoculation with the parasite from human sources is identical with the natural canine disease, while the organism from the canine disease is inoculable to animals, with results similar to those which result from inoculation of the human virus.

"In the present state of knowledge, and lack of absolute proof of the

method of transmission of the disease, it is better to consider all the various systemic diseases in man and dogs as due to one parasite, *Leishmania donovani*. It is hardly necessary to again remark that morphologically (in smears and cultures) the parasites from the various sources are identical.

" Though it is admitted that the human and canine diseases are caused by the same organism, this does not mean that the dog is to be regarded as a reservoir of the virus. Some have maintained that in Italy the disease necessarily passes from dog to man, but so many cases occur which cannot be associated with any infected dog, that it would appear that the infection of the animal is as much an accident as the infection of the human being. Areas occur in which, apparently, only dogs have the disease, while in others only human cases are known. It is claimed, however, by Basile (1916) that in Bordonaro, in Sicily, where a high percentage of naturally infected dogs occurred, the extermination of these has led to an almost complete disappearance of the human disease." (Wenyon.)

Nicolle and other investigators repeatedly examined cats, but never found any sign of the disease in them. Sergent, Lombard, and Quilichini found *Leishmania* in a cat in one case, and this was in a house where a child had leishmaniasis.

In India (Madras) Patton tried to find *Leishmania* in many other different animals, viz., mice, cats and rabbits, goats, monkeys and jackals, but with no result.

CHAPTER VIII.

HERPETOMONIASIS AND LEISHMANIASIS.

A SUMMARY of some of the work performed regarding the connection between herpetomoniasis and leishmaniasis is given below :-

Herpetomonad flagellates have been found by various investigators in insects and in vertebrates. I would refer to the work of Patton, Marzocchi, Nöller, Fantham and Porter, Mackie, Sergent, Shortt and others. They have been found in dog fleas, human fleas, rat fleas, *Anopheles*, culex and bugs among insects, and in mice, lizards and pigeons among vertebrates.

Flagellates of *Leishmania* have been found in the smears taken from certain cases of cutaneous leishmaniasis (Rabagliati, Mongo, Escomel, La Cava).

Many investigators have tried to discover whether it is possible to infect vertebrates with the flagellates which occur naturally in insects. They have arrived at divergent results. Wenyon was really the first to work at this problem (1907). He tried to infect rats with flagellates taken from tabanus, but he met with no success.

A series of experiments performed by Laveran and Franchini, showed the possibility of infecting mice with flagellates found in different fleas, *Anopheles*, *Phlebotomus*, house-flies and *Melophagus*. Subsequently Fantham and Porter, working with the flagellates which occur naturally in *Nepa cinerea*, *Ctenocephalus canis*, *Stratiomyia chamæleon*, *Pediculus vestimenti*, *Gerris paludum* and *Culex pipiens*, were able in a number of experiments to infect fishes, newts, lizards, toads, frogs, mice and birds. On one occasion they succeeded in infecting a snake. In this way they were able to induce acute and chronic herpetomoniasis in vertebrates. Leishmania-like and flagellate forms appeared in their tissues. The organs most affected were the liver, spleen and bone-marrow.

Laveran and Franchini, as well as Fantham and Porter, have described the frequent occurrence of uninucleate parasites in their experimental animals after inoculation. These investigators obtained the same result with feeding as with inoculation experiments.

If these observations receive confirmation then it is possible that leishmaniasis may be an insect-borne herpetomoniasis, and the natural herpetomonads of insects may become pathogenic to man.

Other investigators, however, have repeated these tedious and exhaustive experiments, and Hoare, Shortt and Büchner, among others, have been unable to confirm the above results.

Hoare's experiments, in which he used *Crithidia melophagia* (*Trypanosoma melophagium*), a flagellate employed by Laveran and Franchini in many of their experiments, *Herpetomonas calliphoræ* and *Herpetomonas jaculum* were quite unsuccessful. The vertebrates which he tried to infect were mice, fishes, lizards and frogs.

Shortt also obtained negative results. His experiments were even more exhaustive and complete, because he made cultural investigations in order to determine the presence of infection with *Herpetomonas ctenocephali* and *Herpetomonas lucilæ*. The following is quoted from Shortt's paper :—

"The first point which one has to consider is what constitutes an infection with flagellates. The mere introduction into an experimental animal of a given flagellate and the subsequent recovery of that flagellate before it has had time to be destroyed by the tissues or body fluids of the animal do not constitute proof of an infection. To constitute the latter there should be a true invasion of the organs, tissues, or fluids of the host by the parasite, an invasion which is able to maintain itself for an appreciable period of time with multiplication of the parasite, and which should be demonstrable by the recovery of the parasite by microscopical or cultural methods, preferably the latter, as being less liable to error in the case of positive results. To demonstrate such a true infection by microscopical or cultural methods, these should only be applied some days after the last infecting feed or inoculation, in order to avoid the possible recovery of the parasites which were originally introduced. A large percentage of the experimental animals employed by Laveran and Franchini, and by Fantham and Porter, were found to die as a result of infection with flagellates, and yet in many of these the parasites found post mortem were comparatively few in number. Now, in the majority of protozoal diseases death is not caused except by the presence at some time or other of large numbers of the causative parasite, and we know that in kala-azar the presence in the body of very large numbers of parasites may be quite compatible with the continuance of life for a prolonged period.

"Laveran and Franchini especially, but also Fantham and Porter, make mention of the frequent occurrence in their infected animals of uninucleate forms of the flagellates. What are these uninucleate forms? Do they represent a new stage in the life history of these flagellates, or are they some organism having no connection with the parasites being experimented with? . . . It is true that in cases of true leishmaniasis, and even in insect gut forms, one may see what appear to be uninucleate forms, but it is almost certain that these are merely due to unfavourable orientation of the parasite.

"Franchini and Mantovani's experiments with *Herpetomonas muscæ domesticæ* yielded at least one result which is quite incomprehensible and for which they advance no explanation or even excuse! They state that blood from a rat infected with these herpetomonads yielded in culture 'anaplasma' forms. These forms inoculated into two mice gave rise to a herpetomonad infection. Do these authors suggest that the anaplasmata are stages of herpetomonads?

"The next point one has to consider is the large number of positive results recorded by Laveran and Franchini, and by Fantham and Porter. The latter authors, as already pointed out, got about 85 per cent. of positive results. They used in their experiments, among other animals, martins, frogs, toads, fish, and lizards, all animals whose natural diet is largely if not entirely insectan. Other of their animals, such as sparrows, live largely on insects at certain times of the year, especially during the summer months. If, then, such animals were so easily infected, and so often succumbed to infection, how was it possible to obtain controls, since these also must have swallowed many insects harbouring flagellates? How, indeed, do any of the insectivorous animals, mammalian, avain, reptilian or amphibian, continue to exist?

"The most disappointing feature in the whole record of experimentation with insect flagellates seems to me to be the small use which has been made of modern cultural methods of detecting flagellates of the type used in these experiments.

"In the whole of Fantham and Porter's work there is no single experiment where the positive microscopical findings were controlled by positive cultural results, or even attempts to obtain such, and even Laveran and Franchini until very recently made little use of cultures to confirm their findings. Until 1919 they record no successful culture made from their inoculated animals. In this year they obtained one positive result from a mouse infected by a culture of *Herpetomonas ctenocephali*. In spite of this success they do not seem to have used cultural methods in subsequent experiments, if we except their procedure to get pure cultures of *Herpetomonas jaculum*. These were obtained by inoculating mice intraperitoneally with gut contents of *Nepa cinerea* and then making cultures from the liver. These cultures were presumably obtained for producing infections with pure cultures of *Herpetomonas jaculum*, but no such subsequent experiments were recorded. Hoare, Chatton and Nöller, who used cultural methods to supplement microscopical examinations, got uniformly negative results."

I have already discussed how far experimental evidence goes in favour of fleas transmitting kala azar to man from infected dogs, and have shown the conflicting results obtained by different observers. If, however, the natural flagellates of insects are likely to be converted into *Leishmania*, it then becomes possible that human leishmaniasis, some forms of natural canine leishmaniasis and some other forms of insect flagellosis, may be due to one and the same protozoon.

CHAPTER IX.

CLINICAL VARIETIES OF INTERNAL LEISHMANIASIS OR KALA-AZAR.—COURSE AND SYMPTOMATOLOGY.

Until recently it was thought that Indian kala-azar and Mediterranean kala-azar were two distinct diseases because it was believed that :—

(1) It was possible to recognize differences both in the pathogenic action and biological reactions of the two parasites.

(2) There were definite, though slight, differences in the clinical pictures of the two diseases.

(3) There was a difference between the ages of the patients affected in India and in the Mediterranean countries.

On the other hand, many workers, including Leishman, Nicolle, Pianese, Laveran, Castellani, Rho and others supported Gabbi in the objections which he raised in 1900-10. He showed that :—

(1) The two forms of *Leishmania* (*Leishmania donovani* and *infantum*) have the same morphological, biological and pathogenic characteristics. At the London International Congress of Medicine and Hygiene in 1913, Laveran and Nicolle expressed their agreement with him.

(2) The clinical picture and the pathological anatomy are identical in Indian kala-azar and Mediterranean kala-azar.

Both in the Indian and Mediterranean forms of the disease, complications such as intestinal infection (enteritis, &c.), diseases of the respiratory passages (bronchitis, broncho-pneumonia, pleuritis and tuberculosis), urinary system (nephritis and inflammation of the bladder), and nervous system (meningitis), are observed.

(3) It is not true that in Mediterranean countries only children suffer from internal leishmaniasis. The disease has certainly been observed in adults in these places, although it does not occur so frequently. (Archer, Philipps, Babington, Bassett-Smith, Christomanos, Gabbi, Fucci and Basile, Kalatschnikoff, Gourko, Petrow, Mikoforoff.) In Asia, too, the number of adults infected does not always much exceed the number of children.

In India children frequently become infected, and in a few provinces in China it is children that mostly contract the disease (Cochran, Jerusalemy).

There exists, therefore, no distinction between Indian and Mediterranean leishmaniasis.

Although the parasites of the kala-azars appear to be morphologically identical, and there is much similarity in the symptomatology of the two forms of the disease, yet it is convenient to study them clinically apart.

I. KALA-AZAR OF ADULTS.

The incubation period of kala-azar of adults is said to range from three weeks to several months. In one of Manson's cases the time between the arrival of the patient in an endemic area in India and the onset of the fever was under ten days, or more correctly, as was subsequently pointed out by Low, twenty days. In one of the author's cases the incubation period was not more than one month.

In artificially-infected dogs the disease may remain latent for months, and in monkeys the incubation period is about sixty days. Shortt has recently been able to demonstrate the Leishman-Donovan bodies in smears from the liver in monkeys thirty-nine days after infection.

It was suggested by Sprawson that the incubation period in some of his cases in Mesopotamia was two years, or even more.

The premonitory symptoms of the disease are indefinite. In a large majority of cases the disease begins like typhoid fever. In another class of cases there is history of intermittent attacks of fever, with or without rigor, quotidian for some time and later on becoming irregular. In a third class there is a history of attacks of low fever with slight pyrexia from the beginning. In a fourth class which is, however, rare, there is no previous history of any definite attacks of fever, but the spleen becomes slowly and steadily enlarged. In a fifth class which is also rare, the patient gives a history of gastro-intestinal troubles with dysenteric or diarrhœic attacks, followed by œdema of the legs and attended from time to time with ague-like attacks.

The disease may sometimes be clinically divided into three stages :—

(1) The stage of initial typhoid-like fever.

(2) The stage of secondary low fever or apyrexia.

(3) The stage of cachexia.

Frequently these stages are not sharply marked off from each other, and the typhoid-like pyrexial attacks of the initial period may gradually become less severe until they merge into a period of apyrexia or chronic, low, irregular fever followed by waves of high rises of temperature, and sooner or later cachexia supervenes. Most of the cases begin with symptoms like typhoid fever ; the next in frequency begin like malaria.

Accurate statistics are not available to show the percentage of different modes of commencement of the disease. While in many cases it commences like typhoid or paratyphoid fever, in others it begins as remittent fever with double rises of temperature during twenty-four hours, constituting what has been termed double remittent fever. In others it begins like malarial fever, with intermittent attacks of fever from the very beginning. In these malarial attacks there may be sometimes single rises of temperature during twenty four hours, and sometimes double rises of temperature with single or double intermissions.

Some of the sporadic cases observed by the author in very early stages of the disease in the Calcutta Medical College Hospitals bore a strong resemblance to typhoid fever. The blood showed a partial Widal reaction (1 in 40). In these progressive leucopenia was frequently observed from

week to week. This was much more marked than in an ordinary case of typhoid and was very suggestive of the disease, the diagnosis of which was subsequently confirmed by positive flagellate culture from the blood and by very rapid cure after administration of a few doses of urea stibamine. Double infection of kala-azar and typhoid or paratyphoid fever is not uncommon. In these cases there may be positive Widal reaction (1 in 100 or more), and there is a positive culture for flagellates from the blood, the patient showing definite symptoms and signs of kala-azar after running a course like that of ordinary typhoid or paratyphoid fever. These cases of double infection may lead one to believe that infection with viruses of both the diseases might take place through the intestinal tract. Or is it possible that an attack of typhoid fever precipitates the appearance of symptoms of kala-azar in an individual who may be harbouring the *Leishmania* in a latent stage for some time?

From the very beginning the spleen is enlarged to a greater degree than it is in typhoid or malaria. The liver is also enlarged from the beginning. After the subsidence of the initial stage and the coincident amelioration of the general symptoms, there is a diminution in the size of the spleen and the liver in some cases, while in others they go on increasing steadily. There is either apyrexia during the second period, or probably more frequently the patient gets a low intermittent type of fever of which he is unconscious. Sooner or later after this period of apyrexia there is another febrile attack, and the patient again begins to suffer from some of the types of fever to be presently described which may again be followed by periods of apyrexia. Exacerbations of high fever alternating with apyrexia or low fever occur in the course of the disease. These exacerbations may render it impossible to fix a definite second stage of the disease. After six months to a year the fever may assume low intermittent type which may continue until the end, or it may give way to an attack of the remittent type of fever which may terminate fatally, as has been noted in some cases.

Rigors and sweating have been observed, especially in cases of the malarial type. The terminal stage may be apyrexial, especially in debilitated and emaciated patients.

In the initial stage of the disease anæmia is not a marked symptom, but as the disease becomes established anæmia with a marked and progressive leucopenia is a characteristic phenomenon. Anæmia is not generally so marked as in infantile kala-azar.

Frequently the leucopenia is out of proportion to the anæmia, and a characteristic blood-change may be observed which will be described later on. In some cases, however, the anæmia may be so profound that this characteristic blood-change is not present.

As the disease advances there is a progressive emaciation with œdema of the extremities, the latter sometimes appearing early in the course of the disease. Ascites may be present. There may be hæmorrhages from mucous membranes or under the skin. Epistaxis and bleeding from the gums are common. Hæmatemesis and melæna may take place. Women

frequently suffer from amenorrhœa. There may be menorrhagia. Abortions may occur in pregnant women.

The duration of the disease is variable. It may run an acute, subacute or chronic course. The duration in chronic cases may be two to three years, or even more. In one of the author's cases the patient suffered for more than five years. On the other hand, the disease may run an acute course and terminate fatally in one or two months, or, more rarely, after a few weeks. These acute cases are generally found in places where epidemics occur and during their prevalence.

The cardinal symptoms of a fully established case are : Great enlargement of the spleen and enlargement of the liver, emaciation, cachexia and frequently œdema, irregular pyrexia with tendency towards a double rise during the twenty-four hours, anæmia with progressive leucopenia, the latter being in several cases out of proportion to the former, abdominal symptoms, especially dysentery or diarrhœa, tendency to hæmorrhages and canerum oris.

Symptoms of Kala-azar of Adults considered in Detail.

The types of fever. These are as follows (see charts) :—

(1) Intermittent pyrexia, with or without rigors, of a quotidian type, the paroxysm rarely appearing at the same time every day, but more frequently showing anticipations or retardations (fig. 3).

(2) Irregular intermittent pyrexia, with or without rigors, which may continue for months or pass into any of the other types (fig. 4).

(3) Double quotidian pyrexia with two intermissions during twenty-four hours. There is a rise of temperature towards very early morning, followed by an intermission before 12 a.m., and a second rise in the evening, followed by an intermission before 12 p.m. The heights of the temperature in these two rises are not necessarily equal; they may or may not be accompanied by rigors (fig. 5). In some cases this type of temperature may continue throughout the whole course of the disease.

(4) Double quotidian pyrexia with a single intermission during twenty-four hours. There is a rise of temperature towards very early morning, followed by intermission as in (3). There is a second rise in the evening, which is not followed by an intermission but by a remission before 12 p.m. Both the above types are frequently interchangeable or merge into one another (fig. 6). These double rises of temperature may be continued for months.

(5) Intermittent pyrexia with irregular periods of apyrexia. This is one of the common types of fever in this disease and may be continued for months.

(6) Remittent pyrexia, with a course similar to typhoid (fig. 11). After three or four weeks of illness the patient gets an intermission, and feels better for a variable period until the recurrence of fever. In some cases the remittent type may take on an intermittent type after some days, before a complete apyrexia comes on.

(7) Double remittent pyrexia. There is a rise of temperature towards very early morning, followed by remission before 12 noon. There is a second

rise in the evening, followed again by remission before 12 p.m., but the temperature does not come down to normal. This type, when present, is very suggestive of kala-azar (fig. 7).

(8) Combined intermittent and remittent pyrexia of long duration (fig. 8).

(9) Triple quotidian pyrexia, the rises of temperature taking place at about 3 a.m., 2 p.m. and 8 p.m. (fig. 10).

(10) There may be multiple rises of temperature during twenty-four hours, but this is very rare. In one of the author's cases there were four rises of temperature in one day, followed by five rises the next day (fig. 2).

(11) Finally, there may be indefinite periods of apyrexia (fig. 9).

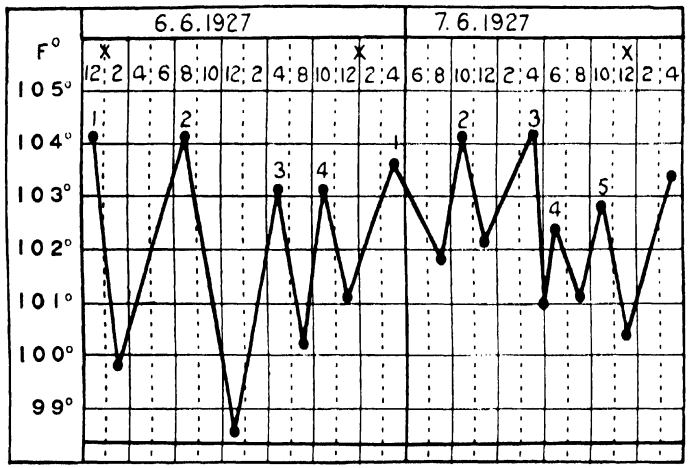


Fig. 2.—Multiple quotidian pyrexia. (Original.)

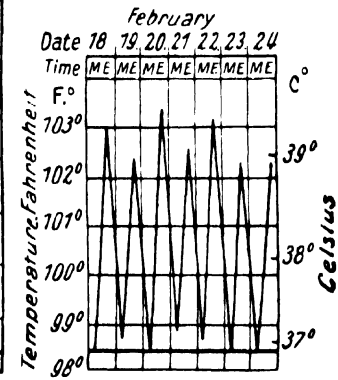


Fig. 3.—Intermittent pyrexia. (Original.)

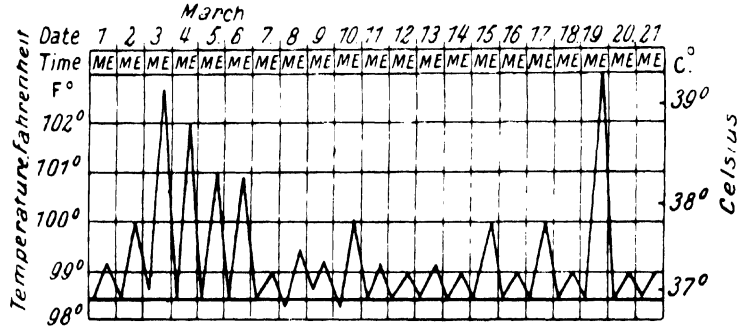


Fig. 4.—Irregular intermittent pyrexia. (Original.)

It should be noted that a double rise of temperature within twenty-four hours with a single or double intermission, continuing for *many months*, does not occur in malaria and is a clear indication of kala-azar.

Chills and Rigors.—Rigors are not uncommon in the type of the disease which resembles malarial fever. There may be two or even three rigors in twenty-four hours, but as a rule there is no periodicity in these attacks. Sometimes there is a chilly sensation at the beginning of the fever.

Nervous System.—Headache is a variable symptom but not so severe as in typhoid, and is more often noted in early than late cases.

Delirium may be present in the initial typhoid-like stage of the disease, and also subsultus tendinum and carphology, but they are rare. Fits resembling

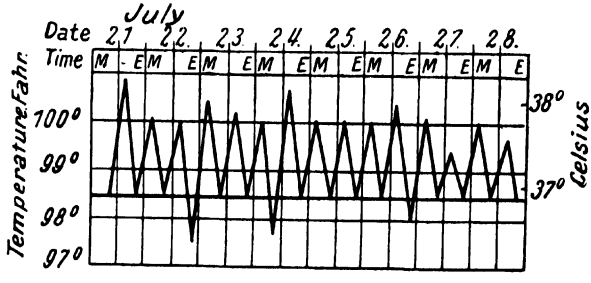


FIG. 5. Double quotidian pyrexia with a double intermission within twenty-four hours. (Original.)

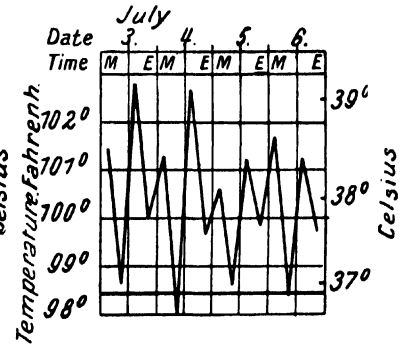


FIG. 6. Double quotidian pyrexia with a single intermission within twenty-four hours. (Original.)

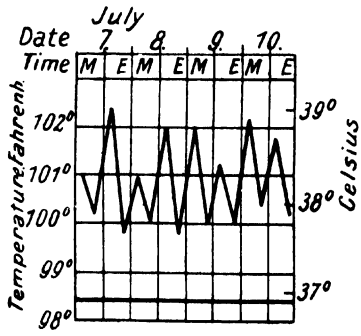


FIG. 7. Double remittent pyrexia. (Original.)

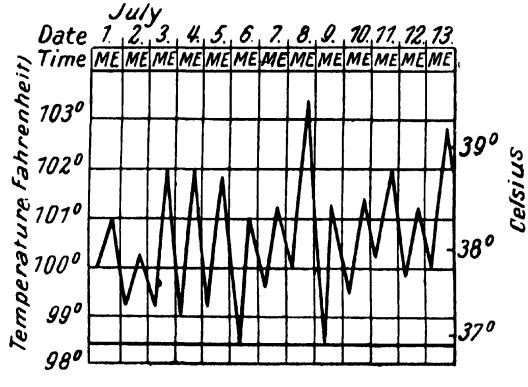


FIG. 8. Combined intermittent and remittent pyrexia. (Original.)

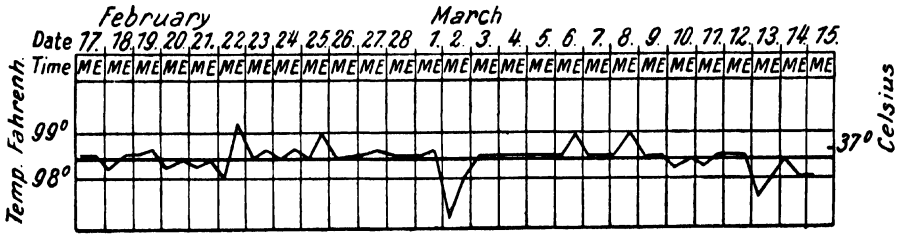


FIG. 9.—An almost apyrexial course of kala-azar. (Original.)

those of tetany, epileptiform convulsions, delirium or coma, are met with occasionally, shortly before death. Generally speaking the mental condition of the patient is quite clear throughout the course of the disease, and even in the high fever of the early stage.

Gastro-intestinal Symptoms.—The tongue is usually clean, even in the early stages of high fever, and the patient has a remarkably good appetite, even when the temperature is high. In some cases, though the appetite is good the digestion is bad, or the patient dislikes his food as soon as he begins to take it. In other cases loss of appetite is a troublesome symptom. Nausea and vomiting are only rarely present, even in the malarial type of the disease. Constipation is more frequent than diarrhœa in the initial stage.

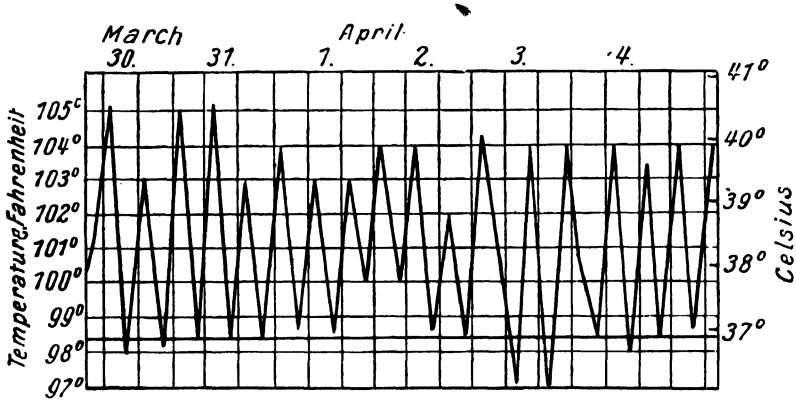


FIG. 10.—Triple quotidian pyrexia. (Original.)

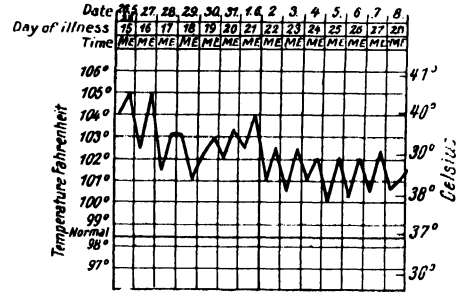


FIG. 11.—Remittent pyrexia. (Original.)

In some cases diarrhœa or dysentery is a persistent symptom throughout the major part of the disease. *Leishmania* dysentery may occur, but is rare. Amœbic or bacillary dysentery is a serious complication in the terminal stages. There may be hæmatemesis and more frequently melæna, apart from any cirrhosis of the liver. Meteorism is not a common symptom of the disease, even during the stage of high remittent fever, and this contrasts with what is observed in typhoid fever.

The Skin.—In many cases the skin assumes a peculiar glistening and stretched appearance, with an earthy-grey colour. Dark pigmentation of the skin is neither so frequent nor so marked as the name of the disease would signify, especially in the sporadic cases that are met with in Bengal. In dark

skinned people the skin of the forehead, the temples, and around the mouth may be more deeply pigmented, but this sign is frequently absent or difficult to recognize. In some cases, however, darkening of the skin is a prominent symptom, especially in the later stage of the disease. This is especially common in Assam. The hair is apt to be dull, dry and brittle, and may fall out. Œdema of the extremities, especially the lower, is sometimes observed early in the course of the disease, and in the late stages there may be general anasarca.

FIG. 12.—Severe emaciation in kala-azar. The swelling of the face indicates commencing cancrum oris.

FIG. 13. Emaciation with marked splenic enlargement in kala-azar.
(Original.)

FIG. 14.—A case of kala-azar with œdema of the face, and general anasarca.

Various forms of skin eruptions are apt to distress the patient. Painful ulcers, wherein the Leishman-Donovan bodies may or may not be found, may be present. There may be hæmorrhages under the skin, especially in the late stages of the disease, and if they occur early in the course of the disease the prognosis is bad. Pemphigoid eruptions may also occur.

General Appearance.—Emaciation is a striking sign early in the course of the disease, though rarely a flabby anæmic appearance may be observed, especially in women. In cases without œdema, the thin face, wasted arms

and legs and prominent ribs are very striking (figs. 12 and 13). Along with the wasting, the abdomen becomes more and more distended, due to the great enlargement of the spleen and the liver, and also to some extent to ascites. The superficial abdominal veins may be dilated, and there is often visible pulsation in the veins of the neck which is sometimes very marked. In cases with œdema, the wasting with prominent abdomen and œdema of the lower extremities gives a characteristic picture of the disease.

Anæmia.—Generally speaking anæmia is not so marked in the early stages of the disease as in malaria. Rarely, however, very severe anæmia may be present, especially when the disease is contracted during pregnancy, or in the puerperal period, or when the disease is complicated with ankylostomiasis or sometimes without any of these complications.

Cardio-vascular Symptoms.—There may be murmurs audible over the heart. Sometimes there may be symptoms of cardiac dilatation. Mackie has laid stress upon tachycardia in the disease, which may be present even when there is no rise of temperature. This tachycardia may be noticeable early in the course of the disease. An unusually slow pulse was observed by the author in some cases a few days before death. A venous pulse is often recognizable in the neck.

Respiratory Symptoms.—Bronchitic symptoms may be present in the early stage of the remittent fever in this disease, but are neither so marked nor so common as in typhoid fever. Towards the termination of the disease bronchitis is frequently present and is sometimes a troublesome symptom. Sometimes there is a troublesome cough due to a relaxed condition of the throat. Œdema of the glottis may occur.

Œdema.—Œdema of the lower extremities is sometimes an early symptom of the disease and is common in its late stages. Sometimes there may be circumscribed œdema. Frequently the patient gives a history of œdema of the lower extremities having been present during some stages of the disease. Rarely there may be œdema of the face and general anasarca (fig. 14).

Urinary Symptoms.—Slight albuminuria is frequently present, and casts are sometimes found in the urine. In a series of twenty cases albumin was present in the urine in fifteen, ranging from a trace to a definite quantity. Hæmaturia and cystitis are rare. The author has observed hæmoglobinuria in two cases in which there were no signs of malaria. Indican may sometimes be present in the urine.

The Spleen.—The spleen is enlarged in almost every case of kala-azar. The great peculiarity in its enlargement is its progressive character. In some cases the spleen may apparently be suddenly enlarged, so that it reaches up to the umbilicus, when the fever has been of a month's duration. The spleen may diminish with the decline of the temperature, but in other cases it continues to enlarge even when the patient has passed into an apyretic stage after the subsidence of the initial remittent rise of temperature. In extremely rare cases there may be little or no enlargement of the spleen, as has been observed in Assam by Dodds Price and others in other endemic areas. Sometimes an attack of melæna or severe diarrhœa is followed by a remarkable diminution in the size of the spleen.

The consistence of the spleen is generally hard, especially in chronic cases, but it is not invariably so, especially in the early stage of the disease. Generally speaking it may be stated that a spleen which is soft or hard and has become much enlarged in a short time is more likely to be due to kala-azar than malaria. Out of a series of 106 sporadic cases in the author's ward it extended from 1 in. to 3 in. in 34 cases, 3 in. to 5 in. in 41 cases and from 5 in. upwards in 31 cases. In this series the greatest enlargement of the spleen was 10 in. below the costal arch. In Rogers' series of the sporadic cases in Calcutta the organ reached up to the level of the navel or below in 85 per cent., while in 10 per cent. it extended to the level of the anterior superior spine of the ileum.

Christophers has described the following types of enlarged spleen met with in endemic areas of kala-azar in India :—

"(1) Large spleens yielding *Leishmania* on puncture.

"(2) Large spleens in which no parasites are found even when many splenic cells are included in the blood drawn from the spleen. Nevertheless, not infrequently in these cases the history and clinical evidence very strongly suggest kala-azar. It is possible that at certain stages of the disease parasites are greatly reduced in numbers or that they are absent from portions of the spleen.

"(3) Very large spleens exactly resembling *Leishmania* spleens, but there is usually a very long history, and the patient, though he may be anæmic, is well nourished and may even be robust. The frequency of this condition among the immigrant coolie population from certain localities, *e.g.*, Arrah, is very noticeable (I think they are probably of malarial origin).

"(4) Very large spleens associated with marked anæmia and often with a considerable amount of œdema of the face and ascites. To superficial examination these cases often simulate kala-azar very closely, but on puncture they yield only malarial parasites."

To the above I would add the following remarks :—

(1) Cases of very enlarged spleen are not infrequently met with in Bengal which do not show the presence of either malarial parasites or *Leishmania*.

(2) Spleens in which the enlargement is quite out of proportion to the duration of the disease is more likely to be due to kala-azar than to malaria.

(3) An enlarged spleen reaching up to the umbilicus after an illness of three or four months is more likely to be due to kala-azar than to malaria.

(4) A spleen which has become much enlarged after an illness of few months and whose consistency is soft is likely to be due to kala-azar.

(5) Some very enlarged spleens may be due to either malaria or kala-azar.

(6) Some very hard spleens may be due to either malaria or kala-azar.

(7) Many cases of moderately enlarged spleens with soft consistency are found to be due to kala-azar.

(8) Rare cases have been met with in which the spleen does not extend more than 2 in. below the costal margin after several months' illness and still they are due to kala-azar.

It would not be correct to say that very hard and very enlarged spleens are mostly due to kala-azar, because they may be due to malaria.

The Liver.—In the majority of cases the liver is enlarged. In chronic cases it is hard on palpation and may undergo cirrhotic changes. In early cases the liver is soft and generally enlarges more quickly than in malaria. Generally speaking the liver is not so enlarged as the spleen, though very rarely the enlargement of the liver may be quite out of proportion to that of the spleen.

Lymphatic Glands.—The lymphatic glands in the axilla and groins may be sometimes enlarged, but not so frequently as in infantile kala-azar.

FIG. 15.—Spleen and liver enlargement in kala-azar. (Original.)

FIG. 16.—Splenic and liver enlargement in kala-azar. (Original.)

The Blood.—As stated before, marked anæmia is not a common symptom in the early stages of the disease. The red blood-corpuscle count varies between 2½ and 4 millions in a majority of cases.

In a series of early cases Rogers found that under one month's illness, in 55 per cent. the red blood-corpuscle count was above 4 millions; between one and six months' illness, in 67 per cent. the red blood-corpuscle count was between 2½ and 4 millions; and over six months' illness, in 28 per cent. the red blood-corpuscle count was below 2½ millions.

Leucopenia is frequently observed in this disease and in many cases early

in its course. The leucocyte count may fall to 700 or 800. Leucopenia is frequently absolute as well as relative. Anæmia and leucopenia are progressive, but the latter progresses more rapidly, so that the ratio of white cells to red cells may be 1 to 1,500, or even less. This relative leucopenia is of great diagnostic importance in the early stage of the disease (Rogers). Rogers points out that in uncomplicated kala-azar the ratio of white blood-corpuscles to red blood-corpuscles is less than 1 to 1,500 in nearly 90 per cent. of the cases, and in a large number of cases the proportion is less than 1 to 2,000, and he concludes that this degree of relative leucopenia is almost absolutely diagnostic of uncomplicated cases of sporadic kala-azar. He also found that in epidemic kala-azar the leucopenia was less marked during high fever than during remissions or low intermittent pyrexia. He lays down as a rule, "that in any cases of fever which may possibly be kala-azar, the finding of less than 1 white to every 1,500 red corpuscles, and still more of greater degree of relative leucopenia, will be almost diagnostic of the disease." This sign may, however, be absent in certain cases, for instance, when anæmia is extreme, as in advanced cases, in pregnancy and the puerperal state, or when complications are present, such as pneumonia, phthisis, cancrum oris, &c., or sometimes even in the absence of any of these factors. The blood-counts of a few cases with marked anæmia are given below :—

(1) Red blood-corpuscles, 1,400,000 ; white blood-corpuscles, 2,170 ; ratio, 1 to 645.

(2) Red blood-corpuscles, 860,000 ; white blood-corpuscles, 2,250 ; ratio, 1 to 382.

(3) Red blood-corpuscles, 1,480,000 ; white blood-corpuscles, 3,870 ; ratio, 1 to 382.

(4) Red blood-corpuscles, 2,300,000 ; white blood-corpuscles, 3,300 ; ratio, 1 to 696.

(5) Red blood-corpuscles, 1,500,000 ; white blood-corpuscles, 2,000 ; ratio, 1 to 750.

These are most puzzling cases, as they fit in with kala-azar as well as with malaria. When the leucocyte count is 5,000 or more, this characteristic relative leucopenia is also absent.

In a successive series of 200 cases in my hospital and private practice, the following was the result of blood examination :—

Red blood-count : In 15 cases above 4 millions, in 121 cases between $2\frac{1}{2}$ and 4 millions, in 49 cases between $1\frac{1}{2}$ and $2\frac{1}{2}$ millions, and in 15 cases below $1\frac{1}{2}$ millions.

White blood-count : In 1 case about 8,000, in 11 cases between 5,000 to 6,000, in 45 cases between 3,000 to 5,000, in 87 cases between 2,000 to 3,000, and in 56 cases below 2,000.

Relative diminution of polymorphonuclear leucocytes is a very constant feature of the disease, and, according to Rogers, this corresponds to a relative increase of the large mononuclears, but, in the writer's experience, relative increase of the lymphocytes is more marked, though it may be less than in infantile kala-azar.

The eosinophils may decrease, according to Rogers, but in many cases they are distinctly increased during treatment with antimony. When the anæmia is severe, poikilocytes, micro- and megalocytes, myelocytes and erythroblasts may be found in the peripheral blood.

The hæmoglobin value of the blood diminishes along with the diminution of the red blood-corpuscles. Rogers holds that the anæmia is of the pernicious type, but frequently it is more of the chlorotic type.

In one series of my cases the hæmoglobin value was as follows :—

In 41 cases it was between 45 to 60 per cent., in 68 cases between 40 to 45 per cent., in 65 cases between 30 to 40 per cent., and in 26 cases below 30 per cent. In some cases it was so low as 15 to 20 per cent.

The average colour index in this series of cases was 0·64, and in about 10 per cent. of them it was nearly equal to the average normal in Indians.

Biochemical Changes in the Blood.—The coagulability of blood is decreased in many cases, and when this decrease is marked there is a tendency to petechial hæmorrhages. There is very little change in the hæmo-salinity of the blood, but its alkalinity may be markedly diminished. In a series of cases, the average basic reactivity was found to be 0·092 normal, as compared with 0·178 normal, in a series of healthy students. In case of Indian kala-azar, the author has found that the relative hæmoglobin value of the resistant erythrocytes during hæmolysis is less than normal.

The experiments in connection with deviation of the complement have not given any reliable results. A limited number of observations were made by me in this direction. In a series of eight cases the reaction was found to be positive in six and negative in two. The diagnosis of each case was made by spleen puncture, and the antigen used was made by the alcoholic extract of fresh spleen of a kala-azar case, made by grinding up one part of the spleen pulp with three parts of a mixture of equal parts of alcohol and 0·85 per cent. NaCl solution, and then heated for an hour at 60° C. The test originally employed by me was Fleming's modification of the Wassermann test. I subsequently tested this reaction according to the original Wassermann method and found that in only one out of four kala-azar cases this reaction was positive. Cornwall found that very careful experiments made with a strong extract of leishmania flagellates and the serum of a kala-azar patient failed to produce any deviation of the complement, pointing to the conclusion that no specific antibodies were present in the blood of a well-marked case of kala-azar.

The Globulin Content of the Blood in Kala-azar.—The author has observed a marked increase in the globulin content of the blood, and probably there is an easily precipitable globulin in kala-azar serum, and these are probably responsible for the globulin tests of kala-azar described by me, and the hæmolytic and aldehyde tests of others. I have separated a globulin from kala-azar serum which possesses marked anti-complementary properties.

The following globulin tests are described :—

(1) Globulin ring test. The serum of the patient is diluted ten to twenty times with normal saline in a test-tube, and then a small amount of distilled

water is gently poured over the serum—a distinct white ring forms over the surface of the serum, similar to that which is observed in the ring test for albumin with nitric acid.

(2) Globulin precipitation test. When the serum is diluted in a test-tube with two to three parts of distilled water, a white precipitate forms. Upon this depends the *hæmolytic test*, which consists of adding one part of blood to two or three parts of distilled water in a miniature test-tube or a Gower's hæmoglobinometer tube. A turbidity forms which consists partly of the precipitated globulin, partly of the stroma of red cells entangling the precipitated globulin and partly of red cells not hæmolysed due to the globulin precipitated on their bodies mechanically preventing their solution. The precipitate settles down at the bottom of the tube after some time.

(3) Globulin opacity test. One part of serum is mixed with six parts of distilled water, and the precipitated globulin obtained thereby is poured into a graduated cylinder, the diameter of which is 1 in. and which contains some black spots at the bottom. More and more of the fluid containing the precipitated globulin is added until the spots become just invisible to the eye looking down on them. The level of the fluid indicates the amount of globulin present. If the level is 1·25 in. or less then it may be regarded as fairly diagnostic of kala-azar.

(4) The aldehyde test. One cubic centimetre of clear serum is placed in a test-tube of $\frac{1}{2}$ in. diameter and 3 in. long, one drop of 30 per cent. formaldehyde (commercial formol) is added to it and the serum is well shaken and placed on a stand. If within a minute or two the serum becomes "set," and then becomes absolutely solid and opaque within three to twenty minutes, it is claimed to be diagnostic of kala-azar. The test is, however, negative in many undoubted cases of kala-azar, especially in the early stages. The turbidity and opacity are also variable in many cases of undoubted kala-azar, as well as in other diseases. Cases which are not kala-azar may be found in which the degrees of opacity and turbidity may be greater than in a kala-azar case. Besides, the degrees of turbidity and opacity vary to a great extent with the time taken for them to develop.

The globulin opacity test being a quantitative one gives more definite results than any other serum test.

Regarding the presence of *Leishmania* in the peripheral blood, see Chapter on Pathology.

It has been recently claimed by Chopra that when a solution of an organic antimonial of the class of urea stibamine is gently added on to the top of the serum of kala-azar patients in a Dreyer's tube, it gives rise to a precipitate at the junction of the solution with the serum, and this is diagnostic of the disease. Chopra's original test consisted of adding a 4 per cent. solution along the side of the Dreyer's tube. In a later communication he suggests that one or two drops of blood may be mixed with 0·25 c.c. of a 2 per cent. of potassium oxalate solution and the test performed in the same way as with the serum.

More dilute solution of urea stibamine has also been recommended.

It would be interesting to follow the value of this test in other diseases.

It is significant that the test seems to be much allied to the globulin ring test of the author described above.

It was long ago recognized by the author that when distilled water is added on to the serum of a kala-azar patient in a test-tube, a marked precipitate forms at the junction and that, as stated in the globulin ring test, a precipitate forms even when the serum is diluted ten to twenty times with normal saline.

II. INFANTILE KALA-AZAR.

The incubation period of infantile kala-azar is also unknown. Gabbi observed an infant seven months old suffering from it, and, judging from the symptoms, he concluded that it must have been attacked when it was four months old. Similar observations have been made by others. Yakimoff and others found shorter periods of incubation in experimental animals, even less than two weeks.

In some cases the course of the disease can be divided into three stages :—

(1) First stage—lasting from one to three months.

(2) Second stage—characterized by constant fever lasting for several months with progressive enlargement of the spleen and liver, anæmia and a characteristic pallor of the skin.

(3) Third stage—characterized by cachexia, attended with purpuric hæmorrhages, marked anæmia and extreme emaciation. There is frequently high and uninterrupted rise of temperature. In exceptional cases there may be apyrexia a few days before death, which is frequently due to respiratory or gastro-intestinal complications.

In the initial stages there is irregular fever associated with a progressive anæmia. In some cases there may be complete intermission of the fever for two or three days at this stage. The patient is dull and suffers from constipation alternating with diarrhœa.

As the disease progresses, wasting and abdominal distension manifest themselves, and the fever assumes an intermittent or remittent type, and there may be two or three rises of temperature in twenty-four hours, as has been observed by Jemma, Caronia and Nicolle. Generally the fever subsequently assumes a continuous type.

The course of the disease may be acute, subacute or chronic (Jemma). The acute type, which is generally fatal, lasts from one to two months and has occasionally been observed in Italy.

The subacute type lasts from five to twelve months, and death may take place from complications. The chronic type lasts two to three years, and may end in recovery. In endemic localities all cases do not die, as has been observed by Jemma, Spagnolio, Nicolle, Lignos, and others, in the Mediterranean districts, and by Balfour in the Sudan.

There is generally progressive enlargement of the spleen, but Dionisi has observed that in very rare cases this may be entirely absent. The liver is not constantly enlarged.

In some cases the disease remains stationary. The cases that improve become more and more bright, the skin regains its normal colour, the spleen diminishes in size and may come to its normal proportions. Subsequently complete recovery takes place.

In those cases that do not recover there is increase of anæmia and weakness, and the patient becomes more and more listless.

The muscular tissue wastes and loses its tonus. Children, especially in the third stage, look very cachectic and pale with sunken features, weary eyes, very pallid lips, lying immobile and apathetic.

In many children a condition of drowsiness is observed towards the end. Subsequently the patient dies of some complication.

Symptoms of Infantile Kala-azar Considered in Detail.

Temperature.—This is frequently of an irregular type, and in the early stages of the disease is interrupted by periods of apyrexia. Sometimes it is intermittent, becoming remittent with double or triple or multiple rises in twenty-four hours. Subsequently the fever assumes a continuous type without any periods of apyrexia. Occasional attacks of hyperpyrexia may occur, and sometimes the temperature may come down to subnormal (see charts, figs. 17 and 18). In rare cases the pyrexia may be slight.

Chills and Rigors. These occur occasionally at the onset of the disease, the seizures coming on suddenly, as in malaria, and terminating with severe perspiration. As a rule, however, these rigors are absent, and the disease begins very insidiously.

Nervous System.—The mental condition of the child deteriorates as the disease progresses. He will no longer play, and becomes more and more indifferent to his surroundings as the anæmia increases. He loses his sprightliness and always rests in his mother's arms, and sometimes becomes stuporose towards the end.

Gastro-intestinal Symptoms.—Vomiting and diarrhœa, or constipation alternating with diarrhœa, are frequently present and sometimes are the first symptoms that are noticed in this disease. There is a good appetite which may be increased at times. Rarely there may be anorexia. The stools may contain undigested food material with blood and mucus. The motions are very foul. Hæmorrhage from the intestines is rare. There may be terminal dysentery or diarrhœa.

The Skin.—As the disease progresses, the skin loses its elasticity, and assumes a peculiar pale, transparent, wax-like or earthy appearance; sometimes it appears cadaverous-looking. Pin-point hæmorrhages, petechial and purpuric hæmorrhages, are common on the trunk, the abdomen and the extremities. They are probably more common than in Indian kala-azar.

General Condition.—Anæmia is more intense than in adult kala-azar, and this is accompanied by wasting and disappearance of the subcutaneous fat. The prominent ribs, scapulæ and long bones, the protuberant abdomen due to enlargement of the spleen and liver, and ascites, and the enlargement of the superficial abdominal veins present a very characteristic

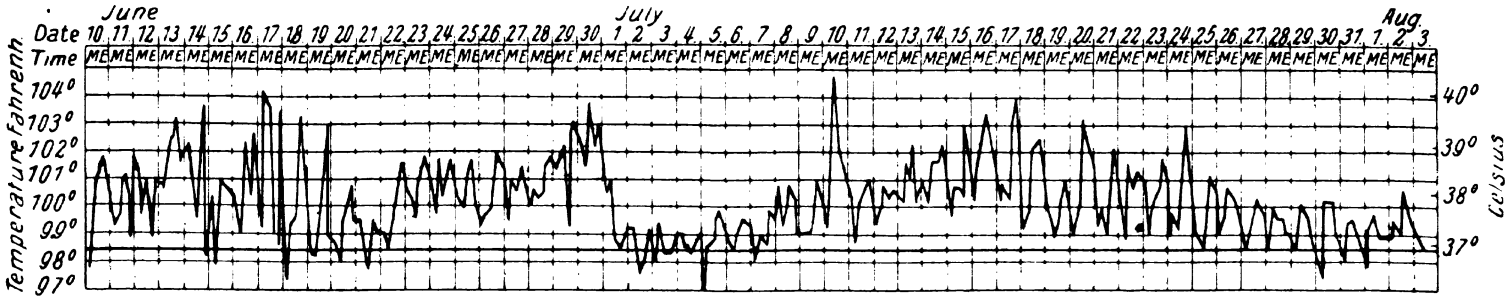


FIG. 17.—Temperature chart of a child with kala-azar. (Nicolle and Ortona.)

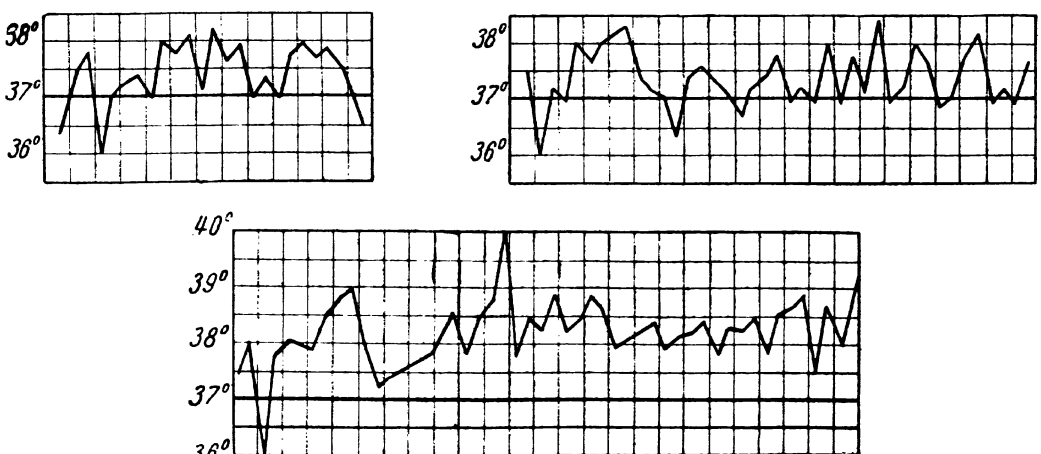


FIG. 18.—Variations in the course of temperature in a case of infantile kala-azar during three months. (Di Cristina.)

picture. Later on œdema of the extremities, the face and the eyelids, shows itself.

Œdema may arise in any stage of the disease, and sometimes it appears suddenly. Early œdema often vanishes suddenly. The œdematous areas may be inflamed and become painful.

There may be visible pulsation of the veins of the neck.

Diminution in Weight.—Progressive from the beginning, and is especially marked towards the end.

Cardio-vascular Symptoms.—The pulse is always very rapid, and its rate out of proportion to the rise of temperature. Tachycardia remains during periods

FIG. 19. Kala-azar in a child aged 14 years. (Original.)

of apyrexia, and during the fever the rate may be 150-160. Hæmic bruits may be present in the heart.

Respiratory Symptoms.—There may be bronchitic symptoms and dyspnœa on exertion. There may be cyanosis and dangerous asphyxia, sometimes due to œdema of the glottis. Lobar pneumonia, broncho-pneumonia or pleurisy may be sometimes observed and especially towards the end.

Urinary Symptoms.—The urine usually remains normal, but there may be slight albuminuria, or diminution in the output. Lipuria may be present (Franchini and Longo). Petrone and Cannata found the diazo reaction present. Rarely there may be cystitis. Scordo found no abnormality in the ratio between the chlorides and nitrogen.

The Spleen.—The spleen becomes progressively enlarged, till it fills the left side of the abdomen, but there is no constant relationship between the size of the spleen and the severity and progress of the disease, as has been stated erroneously. In the first stage the spleen is soft, and it begins to harden in the second and third stages. The spleen may diminish in size towards the end, especially after profuse diarrhœa. The absence of enlargement of the spleen is very rare. The patient may complain of pain in the spleen and occasionally small supernumerary spleens may be present.

The Liver.—It is generally enlarged, but it bears no recognizable relationship to the enlargement of the spleen. Its edge is regular, sharply defined and firm. Some observers lay greater stress upon the increasing hardness of the liver than upon its enlargement. Hepatoptosis is very rare (Gabbi). Sometimes there is no enlargement of the liver.

FIG. 20.—A case of infantile kala-azar. (Original.)

The Lymphatic Glands.—The glands in the neck, axilla and groin, frequently undergo painless enlargement, according to some observers, while others state that they are as a rule unchanged. Jemma and di Cristina described a case in which micropolyadenitis occurred. Feletti, Cortesi, Levy, Spagnolio and Caristo observed simple lymphadenitis in the region of the neck, axilla and groin. Cochran and Neumann, as well as Spagnolio, have found *Leishmania* present in these glands.

The Blood.—The blood-changes are somewhat similar to those observed in kala-azar of adults. But the relative leucopenia is not so pronounced. In the Sudan variety of the disease, diminution of the eosinophils has been observed. The blood may rarely show the presence of microcytes, macrocytes and poikilocytes, and, still more rarely, normoblasts. The hæmoglobin content of the blood may be as low as 10 per cent. and the colour index is less than normal. Cannata found the resistance of the red corpuscles diminished, while Abate observed increase of maximum and minimum resistance. The red corpuscles diminish in number as the disease progresses. As regards the leucocytes, Cannata found in 31·1 per cent. the leucocyte count was normal or nearly

normal, in 62·2 per cent. there was leucopenia, and in 6·7 per cent. there was slight leucocytosis.

Feletti and Longo, in Catania, and Kardematis, in Greece, and Pittaluga, in Spain, always found leucopenia. Generally there is relative increase of the large mononuclears and lymphocytes at the expense of the polymorphonuclears. The presence of lipemia, noted by Longo, has been confirmed by Gabbi.

As regards the biochemical changes in the blood, Milo, in Messina, and others have confirmed the author's observations on the globulin precipitation test in serum of adult kala-azar.

Pavoni found that the complement-deviation reaction in cases of kala-azar running the usual course of the disease was nearly always negative. In some cases that recovered, the reaction was positive as also in chronic cases, though it might disappear suddenly if the patient suddenly became worse. The reaction for deviation of the complement was tried by Cristina and Caronia in eighty-eight cases of infantile kala-azar, with partially or definitely positive results in thirteen cases. In those cases in which a positive result was obtained the malady had only been of short duration, suggesting that there was an immunity reaction early in the disease. They also tried the complement-deviation reaction by the method suggested by Ascolti. It depends upon the fact that a subject who has been submitted to the action of an antigen without having reacted in the production of specific antibodies may be artificially stimulated with the small doses of the same antigen (killed cultures, toxins, &c.), so that he acquires the power of forming these antibodies (Ascolti's allergic sero-diagnosis). Patients, suffering from kala-azar, in whose blood no antibodies could be detected were treated with nucleoproteids extracted from cultures of *Leishmania*. Their blood showed the reaction for the deviation of the complement and agglutination on the fifth or sixth day.

Di Cristina and Caronia, as a result of their own observations and those of other observers, conclude that agglutinins and specific precipitins only rarely are produced in any quantity in the blood as the result of the natural *Leishmania* infection, but that it is possible to produce them artificially by the injection of killed parasites or of nucleo-proteids extracted from these. They hold that the *Leishmania* act like other micro-organisms, in that they stimulate the body by means of toxins and endotoxins to the production of antibodies. In the cases in which a positive result was obtained, the malady had only been of short duration, suggesting that there was an immunity reaction early in the disease only. In one case this disappeared after fifteen days. The reader is referred to Noguchi's experiments on serological reactions (p. 35).

Scordo found that the serum of kala-azar patients had some agglutinating action on cultures of *Leishmania*, and that this action was more on the cultures of human than of canine origin. Bandi has observed that the repeated intravenous injection of rabbits with cultures of *Leishmania* of human or canine origin causes the appearance in the blood of substances capable of agglutinating the cultural forms. The serum of rabbits injected with cultures

of canine *Leishmania* agglutinated cultures both of canine *Leishmania* and *Leishmania infantum* in dilutions of 1 in 160. Their sera further produced agglutination of cultures of *Leishmania tropica* in a dilution of 1 in 70. Montenegro reports that intradermal injection of alkaline extract of *Leishmania* given to patients with leishmaniasis produces a positive reaction in more than 80 per cent. of cases, which, when typical, is diagnostic.

Caronia holds that the complement power of the blood is increased in cases of kala-azar, being more marked in the later stages of the disease. In this connection the author would refer to his observations that the serum of kala-azar patients contains a peculiar globulin-like substance which possesses a marked anti-complementary property when mixed with a hæmolytic system. The author's observations on Indian kala-azar differ from those of Caronia

CHAPTER X.

COMPLICATIONS.

I. IN THE ADULT FORM.

CANCRUM ORIS is common, and is a most serious complication. There may be extensive sloughing of the cheeks, and destruction of the muscles and bones of the face (see fig. 21). The sloughing process may also extend upwards and destroy the eyeball. Septic absorption may take place into the vessels of the brain, giving rise to cerebral complications, such as meningitis, &c., followed by convulsions, coma and death. In other cases, the sloughing process may extend to a large artery and severe hæmorrhage may follow. Death may take place from hæmorrhage from the facial or the lingual artery. In rare cases, sloughing may start from the soft palate, the pharynx, or from under the tongue. The tonsils may slough off. Bleeding from the gums, ulcerative stomatitis, and pyorrhœa alveolaris are often observed. Septic conditions such as ischio-rectal abscess, mastoid abscess, otitis media, gangrene of the vulva, as well as sloughing and septic dysentery, may occur. There is a tendency to hæmorrhage from different parts of the body, as shown by petechiæ or large purpuric patches. These are serious complications, and appear more in the late than in the early stages of the disease. Hæmatemesis and melæna may occur and sometimes herald the end. These may be followed by a striking diminution in the size of the spleen. Melæna may occur with or without any dysentery. Epistaxis may be severe and threaten life. It may be present either early or late in the disease. There may be cerebral hæmorrhage and death may follow.

Gastro-intestinal complications, such as diarrhœa, bacillary or amœbic dysentery, are often terminal complications. Cases have been described in which dysenteric symptoms due to *Leishmania* closely allied to those of true dysentery may appear during the course of the disease. Such leishmania dysentery must be very rare, and generally the dysentery is of the bacillary or amœbic type. Generally speaking, dysentery is a serious complication, and if it tends to be chronic it frequently ends fatally. Septic dysentery is almost always fatal. If dysentery is associated with marked œdema and anæmia, it is then frequently fatal. Very rarely death may take place from perforation of the bowels.

Bronchitis is frequently present when there is marked œdema. A relaxed condition of the throat may give rise to a troublesome cough. Broncho-pneumonia, lobar pneumonia, pleurisy, empyema and tuberculosis, may also

complicate the disease. If pneumonia is associated with cancrum oris or a septic condition of the mouth, then it is very likely to end fatally. One case of the author had hæmothorax, and sixty ounces of bloody fluid were removed from the left pleural cavity.

Reference has already been made to tachycardia. More rarely there may be marked bradycardia a few days before death. Dilatation of the heart may be present. Jaundice and ascites due to a cirrhotic condition of the liver, or pressure of cells loaded with *Leishmania* upon the bile or the portal capillaries may be present. Liver abscess is rare. Perihepatitis, perisplenitis, infarcts in the spleen and its rupture have been noted. The author has seen extensive hæmorrhage under the capsule of the spleen, giving rise to a hæmatoma.

FIG. 21.—A case of extensive cancrum oris in adult kala-azar, cured by urea stibamine. (Original.)

Severe anæmia, general anasarca, albuminuria, and true nephritis may be present.

Ankylostomiasis, typhoid fever, and *Bacillus coli* infection may complicate the disease. Malaria may complicate the disease, but not so often as one would expect from the common endemicity of the two diseases in many places. This is partly to be attributed to the fact that most cases come for treatment after having taken quinine, having been primarily diagnosed as cases of malaria. In hospital practice malaria is rarely found as a complication of kala-azar.

In acute cases, typhoid symptoms, subsultus tendinum, delirium and coma, are occasionally present. These may also be present in some chronic cases shortly before death. Tetany, epileptiform fits, retinal and meningeal hæmorrhages are unusual complications.

Dropsy is sometimes found to be a very marked symptom of the disease and is frequently associated with much anæmia. Cases with severe anæmia and marked dropsy are generally very resistant to treatment.

Skin diseases and ulcers in the skin frequently arise in chronic cases.

II. IN THE INFANTILE FORM.

Hæmorrhages may also occur in this form of the disease, such as severe epistaxis, which may be fatal, hæmorrhage from the gums, petechiæ and purpuric patches. Melæna and hæmatemesis are not common. Bleeding from the gums is common. There may be complications of stomatitis *catarrhalis* and *ulcerosa* and noma. The former makes its first appearance upon the lips, and subsequently affects the cheeks or the gums. It may be present either in a slight (*forma catarrhalis*) or severe form (*forma ulcerosa*) and resemble true noma. Noma has been observed in Tunis by Nicolle, in Italy by Gabbi, Longo and Feletti, in Greece by Lignos and Christomonos, and in Malta by Critien.

Critien observed cases of cancrum oris in which the course of development varied from three to six weeks. As long as the condition was maintained, there was practically no pain. The affected mucous membrane had a greenish-black superficial appearance; it secreted a malodorous mucilaginous fluid, which dripped away at the corners of the mouth. In one case, observed by Longo, the infective process spread to the palate and nasal bones and destroyed both. *Leishmania* have never been found in the secretion, except in one of Gabbi's cases, in which instance he thought that they probably had come from the blood, because they were found in a moribund child.

Septic infections, mastoid abscesses, otitis media and deafness, may complicate the disease.

Respiratory complications are not unusual. There may be bronchitis, lobar pneumonia, broncho-pneumonia, pleuritis and tuberculosis. Sometimes there develop fatal dyspnœa and cyanosis, due probably to œdema of the glottis. Albuminuria, true nephritis, hæmaturia and lipuria may also complicate the disease. Malaria and ankylostomiasis may be associated with infantile kala-azar. Obstinate diarrhœa, entero-colitis and dysentery may be dangerous complications, and lead to death. In many cases these intestinal disorders are due to errors in diet. There may be cerebral hæmorrhage and spinal meningitis. Skin eruptions may be present, e.g., pemphigus (Nicolle).

CHAPTER XI.

PROGNOSIS.

THE prognosis of kala-azar in cases of untreated adults is very grave. Before treatment with antimony had been introduced about 90 to 95 per cent., or even more, of the cases died. In Assam "it is a well-known fact that before any treatment for kala-azar was known, at least 90 per cent. of those attacked succumbed to the disease" (Murison). A very low leucocyte count, marked diminution of the polymorphonuclears, and severe anæmia indicate an unfavourable prognosis. Complications influence the prognosis, especially cancrum oris, dysentery, diarrhœa and hæmorrhagic tendencies which are very serious. Cases of cancrum oris without reactive leucocytosis invariably terminate fatally, while those with a well-marked reactive leucocytosis may recover both from kala-azar as well as cancrum oris. Other complications that influence the prognosis are pneumonia, nephritis, cirrhosis of the liver, cardiac dilatation, and general anasarca. If the patient recovers from an attack of pneumonia complicating the disease, then he may also recover from kala-azar, especially if a well-marked leucocytosis develops during the attack of pneumonia.

There are rare cases that are resistant to antimony treatment, and their prognosis is bad.

In infantile kala-azar, 14 to 24 per cent. of the cases have been recorded by different observers as undergoing spontaneous cure. Here, too, complications influence the prognosis.

Jemma has observed that the chronic type of the disease, lasting from two to three years, may end in complete recovery. Noma, dysentery, obstinate diarrhœa, hæmorrhagic tendency, and œdema of the glottis are the most dangerous complications. The more marked the leucopenia, and the smaller the number of polymorphonuclears, the worse is the prognosis.

In the Sudan the percentage of spontaneous recoveries is very small. It has been noted that a reappearance or increase of the eosinophils in the blood is a favourable sign. Increase of eosinophils occurs in cases of kala-azar of adults that are improving under antimonial treatment.

Since the introduction of the antimonial treatment of the disease no case should be regarded as hopeless, unless there is a fatal complication.

CHAPTER XII.

DIAGNOSIS AND DIFFERENTIAL DIAGNOSIS.

I. DIAGNOSIS.

IN areas where kala-azar of adults is endemic, any doubt arising as to the correct diagnosis of the condition can be settled if the following symptoms are present :—

A double rise of temperature persisting for a long time ; progressive, rapid and marked enlargement of the spleen ; increasing emaciation and loss of weight ; progressive anæmia and progressive leucopenia ; early œdema and hæmorrhages ; a peculiar, somewhat shiny appearance of the skin ; and the failure of quinine in checking the fever.

If the ratio of the white to the red corpuscles in the blood is less than 1 to 1,500, then the disease is very likely to be kala-azar. Unfortunately this relative leucopenia is not always present. The globulin tests are of diagnostic value when positive. Brahmachari considers that the globulin opacity test is of great value. The formaldehyde test may be negative in early cases of undoubted kala-azar, while it may be positive in cases that are not kala-azar. Chopra's recent urea stibamine may also be tried. Di Cristina and Caronia's anaphylactic test described below does not appear to have been tried in kala-azar of adults.

"Wagener (1923) has shown that the injection of alkaline extracts of *Leishmania* from cultures into the skin of rabbits previously rendered sensitive by injections of cultural forms of *Leishmania* produces a local reaction in the form of an erythematous papule, which reaches its height in forty-eight hours, and persists from three to five days. The antigen can be prepared from both *Leishmania tropica* and *Leishmania donovani*, as it is not specific for either parasite. If these results are confirmed, the reaction may be of use for diagnostic purposes" (Wenyon's PROTOZOOLOGY).

Infantile kala-azar presents a similar clinical picture. The differences which occur are dependent on the age of the patient. The temperature is more irregular, and the characteristic leucopenia of kala-azar in adults may be absent. The enlargement of the spleen and the liver, with distended superficial veins in fairly advanced cases, the irregular fever, pallor and anæmia, occurring in a child in an endemic area, make the diagnosis easier. Milo has used the globulin precipitation test and has invariably obtained positive results. The other biochemical tests, e.g., the globulin opacity and the formaldehyde tests, should be tried. Di Cristina and Caronia have

described a method of diagnosing infantile kala-azar by what they call the anaphylactic test. It is described as follows :—

2-3 c.c. of serum from a patient is mixed with 2 c.c. of culture of *Leishmania* on N N N medium. The mixture is kept at 37° C. for one hour, and then on ice for twenty-four hours. It is injected intravenously into a guinea-pig which has been immunized against culture of *Leishmania* by repeated subcutaneous injections. Reaction as shown by tremor, dyspnœa, paralysis of the sphincters, and even death, may be observed in cases of kala-azar, while with the serum of healthy children no such reaction is shown.

The presence of Leishman-Donovan bodies in the spleen, the liver, the bone-marrow, or in the peripheral blood, clinches the diagnosis of the disease. Careful examination of the blood-smears from the peripheral blood, especially at the margins of the smears where the leucocytes collect in largest numbers, may show the presence of *Leishmania*. It may be advantageous to constrict the finger by a ligature for some time before the blood is taken from it. Unfortunately the method is tedious, because often only very few parasites are present in the peripheral blood, and they may easily be overlooked. As the parasites are present inside the large mononuclear or polymorphonuclear leucocytes, these cells should be carefully examined. The blood may be citrated with 4 per cent. solution of sodium citrate and centrifuged. The deposit in the upper layer of the sediment, which consists mostly of leucocytes, may be examined for the presence of *Leishmania*.

Knowles and Gupta, using the following method, were able to show the presence of parasites in the peripheral blood in 67 per cent. of the cases they investigated. Four large drops of blood are placed one in each corner of a half-inch square on a glass slide, and by means of a needle or glass rod they are spread thickly and evenly. The slide is then dried at room temperature for two hours, or in an incubator at 37° C. for one hour. The haemoglobin is removed from the smear by using a mixture of four parts of $2\frac{1}{2}$ per cent. solution of acetic acid in distilled water, and one part of a 2 per cent. solution of crystalline tartaric acid in distilled water. The smear is then fixed with methyl alcohol and, after thorough washing with distilled water, is stained. The *Leishmania* are found inside the large hyaline mononuclear or polymorphonuclear leucocytes. For other methods of preparing thick films see Appendix and Addendum.

In the author's experience, the smears made after puncture or excision of the lymphatic glands, as suggested by Cochran, have given negative results. It has also been suggested that the contents of artificial pustules of the skin made by irritation and the leucocytes present in them, and scrapings from ulcers, should be examined. The results have been unsatisfactory in the author's hands. Bone puncture may give more satisfactory results, but it is not always easy and is rather painful, and a procedure not devoid of danger. The head of the tibia, the lower end of the femur, a rib or the sternum may be perforated and the marrow removed and examined for *Leishmania*. The removal of the bone-marrow may be facilitated by the

use of a trochar and cannula aided by an aspirator. Liver puncture has been advocated and is considered to be less dangerous than spleen puncture. But the results are not always positive, even in cases of kala-azar.

The surest method of finding the *Leishmania* in the smears of tissues is by spleen puncture.

A small all-glass hypodermic syringe of 1 or 2 c.c. capacity with a fine hypodermic needle is used to enable one to draw enough of the spleen pulp to show the presence of parasites. It is advisable to give the patient 10 to 15 grains of calcium chloride half an hour before and after the operation. The needle must be perfectly aseptic and dry, and the operation gently performed. The needle and the syringe, after being sterilized in oil or boiling water, are dried with absolute alcohol which is afterwards blown out as much as possible with the aid of the piston, the last traces being allowed to evaporate. If there is any trace of water in the needle, then the parasites will burst and be distorted and thereby rendered unrecognizable, and the presence of alcohol may coagulate the blood and thereby block the needle. The patient should not speak or breathe after the needle has pierced the skin and has penetrated the spleen. The patient should lie on his back, and the needle should be inserted more or less perpendicularly. The insertion is made at a point where the spleen seems to be in greatest contact with the abdominal wall. It is always desirable to fix the skin of the spot to be punctured with two fingers of one hand. If much blood is drawn by the syringe, it is advisable to exert digital pressure on the punctured spot for one-half to two hours, and to keep the patient absolutely quiet.

Opinions differ as to the danger of spleen puncture. However simple the operation, and however slight the danger may be, it certainly does exist. In the early stages, when the spleen is soft and very vascular and the capsule is thin, the danger of hæmorrhage is great. On the other hand, in the later stages of the disease the spleen is more friable, and a puncture may make a rent which may not close after removal of the needle. Besides, in this stage the coagulability of the blood is sometimes very markedly diminished, and this may increase the tendency to hæmorrhage. If, however, the precautions described are followed and one uses a fine needle and does not attempt to draw large quantities of blood, then the dangers of the operation can be reduced to a minimum. In cases where there is a hæmorrhagic tendency and the coagulability of the blood is diminished, the operation should not be performed. The latter condition can be recognized when the coagulation time for the peripheral blood of the patient is longer than five minutes. Other contra-indications are the presence of severe anæmia, the red blood-corpuscles being less than two millions per cubic millimetre, severe cough, diarrhœa, nausea, vomiting, presence of much ascites, or any condition in which there is much movement of the diaphragm. The possibility of the case being leukæmia or typhoid fever should also be borne in mind. In any case the operation should not be performed unless it is absolutely necessary.

The absence of *Leishmania* from spleen puncture does not absolutely exclude kala-azar. Christophers thinks that in certain stages of the disease

they are much reduced in number, and may be absent from certain portions of the spleen. It may happen that microscopic examination of spleen puncture material does not reveal any *Leishmania*, but a culture prepared from spleen tissue will show their presence.

The culture of the parasites from the peripheral blood should always be undertaken in suspected cases, if there are facilities for doing so. This method was successfully carried out by Novy, Mayer and Werner, Wenyon, Cornwall and others. Row has elaborated an "intensive method" of cultivating the parasites. A few drops of blood from the finger are introduced directly into 15 to 20 c.c. of citrated saline solution before any clotting has taken place. This diluted blood is then centrifugalized and the sediment of corpuscles planted directly into a suitable medium, e.g., N N N medium. In this way a growth of flagellates may be obtained in about six days. The following modification of his method gives very good results :—

About a quarter of a cubic centimetre of blood from a vein at the bend of the elbow is put into 20 c.c. of citrated salt solution (physiological saline containing 1·5 per cent. sodium citrate); the mixture is shaken gently and allowed to stand over-night in the cool incubator. As soon as the corpuscles have settled to the bottom of the tube, the supernatant fluid is poured off, and the corpuscles are pipetted off with a capillary pipette and transferred into the water of condensation at the bottom of N N N tubes, which are then incubated at 22° C. In the case of splenic blood, the syringe is filled with a few drops of the citrate solution mentioned above and the spleen punctured. The blood drawn is then mixed with the citrated saline inside the syringe, transferred to the N N N medium and incubated in the same way as the peripheral blood. Blood from a finger may be taken for culture instead of the blood from a vein.

N.B.—To ensure a successful result, strict aseptic precautions are necessary, as slight bacterial contamination kills the *Leishmania*, though they have been found to grow luxuriantly with fungi.

II. DIFFERENTIAL DIAGNOSIS.

In the early stages, kala-azar may be mistaken for *malaria*, *typhoid* or *paratyphoid fevers*, or *Bacillus coli* infection. Cases that begin with rigor and fever of an intermittent nature are likely to be mistaken for acute malaria. On the other hand, malignant tertian fever, in which the fever may be irregular and sometimes assumes a remittent type and may not be associated with rigor, is likely to be mistaken for kala-azar.

In such cases the blood should be examined for malarial parasites and pigment-bearing leucocytes, and the effect of quinine must be tried. If the fever resists quinine administered orally for a week, in doses of 0·65 to 1·0 grm. (10 to 15 gr.) per day, and has also resisted intramuscular and intravenous injections of quinine in 0·65 grm. (10 gr.) doses for a week, then it may be concluded that the infection is not malarial, and other diseases, among which one may include kala-azar, in an endemic area, should be suspected.

It must not be forgotten that in some rare cases there may be a double rise of temperature and marked leucopenia in malaria. Marked periodicity of the paroxysms of fever, and heavy perspiration towards the end, are more common in malaria than in kala-azar. Bleeding from the gums and epistaxis are more common in early cases of kala-azar than in malaria. A voracious appetite is a symptom in favour of kala-azar.

Generally speaking, in the early stages the anæmia is greater in malaria than in adult kala-azar, while on the other hand, marked leucopenia occurs more frequently in kala-azar. The characteristic relative leucopenia of the disease, if present, is very helpful in diagnosis. On the other hand, the blood-picture may be somewhat as follows: Hæmoglobin, 55 per cent.; red blood-corpuscles, 3,200,000; white blood-corpuscles, 5,000. Such a blood-picture may fit very well with a case of early kala-azar just as much as it may fit with malaria. The globulin tests should help in the diagnosis. The smears from spleen puncture material should be examined for Leishman-Donovan bodies, a peripheral blood-culture and spleen blood-culture should be made for flagellates, if there are facilities.

Careful examination of the temperature chart should be made. A marked double rise of temperature continued for days, with tachycardia, which is persistent during periods of apyrexia, points to kala-azar. Early œdema associated with slight enlargement of the spleen, in the absence of nephritis and anæmia, is more frequent in kala-azar than in malaria.

Early and acute cases may be mistaken for *typhoid fever*. The characteristic step-ladder-like temperature may be absent in a case of typhoid, just as the double rise of temperature may be absent in a case of kala-azar. Double rises of temperature may rarely be present in cases of typhoid, especially in children.

Generally speaking, there is no great disturbance of the nervous system in kala-azar. Leucopenia and relative increase of lymphocytes may be present in both the diseases. A positive Widal reaction in dilutions of 1 in 100 goes much in favour of typhoid. It must, however, be remembered that the blood in the early typhoidal stage of Indian kala-azar may give a partial or rarely a completely positive Widal reaction, probably due to a double infection. In the case of infantile kala-azar, Cannata never found the reaction positive. On the other hand, a positive Widal reaction may be absent, though rarely in undoubted cases of typhoid. The enlargement of the spleen is more marked in kala-azar. In doubtful cases blood-cultures help to confirm a diagnosis.

Paratyphoid fevers are frequently mistaken for kala-azar. Severe constitutional symptoms are likely to be absent in both these diseases, and the spleen may be considerably enlarged in them. Shivering may be present in early stages of both the diseases.

Although paratyphoid fever generally lasts for a fortnight, it sometimes lasts for two or three months, or even longer, and may be thus mistaken for kala-azar. Positive Widal reaction for paratyphoid (A or B) goes in favour of paratyphoid fever.

Another disease for which kala-azar is likely to be mistaken in its early

stage is *Bacillus coli infection*. The irregularity of the temperature curve, and the occurrence of rigors with high rises of temperature, sometimes more than once during twenty-four hours, may take place in both diseases. The difficulty in diagnosis is specially likely to happen during pregnancy or in the puerperal state, in which the characteristic relative leucopenia of kala-azar may be absent. The presence of marked leucocytosis goes in favour of *Bacillus coli* infection. In doubtful cases the spleen should be punctured or a blood-culture made for flagellates, in order to confirm the diagnosis.

Acute tuberculosis may be associated with leucopenia, enlargement of the spleen, and remittent type of fever, and there may be no physical signs of tuberculosis in the lungs. In acute tuberculosis, dyspnœa, cyanosis and choroidal tubercles are likely to be present. Generally speaking, symptoms of tuberculous meningitis show themselves, and cerebral symptoms are more marked in this disease than in kala-azar.

In chronic cases of kala-azar, low intermittent fever may lead one to suspect *chronic tuberculosis*, especially in a patient who is markedly wasted, and has been suffering from obstinate diarrhœa or dysentery. In addition the bronchitic symptoms, which are often present in the later stage of kala-azar, may suggest chronic tuberculosis, or kala-azar complicated by tuberculosis. Tuberculosis in which the spleen is enlarged may be mistaken for kala-azar.

Tuberculous infection should be suspected in a case of kala-azar, if during antimony treatment the fever changes to a hectic type and there is no improvement in the patient's general condition, in spite of disappearance of leucopenia and appearance of leucocytosis and subsidence of splenic and hepatic enlargements. In such cases a careful examination for a tubercular focus in the lungs or elsewhere should be made.

In chronic cases the temperature curve of *Malta fever* may resemble that of kala-azar, so that there may be difficulty in the diagnosis in localities where both diseases occur. Generally speaking, the spleen in Malta fever is not so much enlarged as in kala-azar, and the liver is hardly enlarged in it. Constipation and heavy sweats are fairly constant symptoms in Malta fever, while hæmorrhages, œdema, pigmentation of the skin and leucopenia are absent in this disease. There may be slight leucocytosis. The serum test will be of great help in diagnosis, and the intimate relationship of the disease with the use of infected goat's milk may be of help in establishing the diagnosis. Anæmia is more marked in kala-azar. Malta fever runs a much milder course than kala-azar.

Trypanosomiasis and kala-azar may be difficult to differentiate in those rare places where the two diseases occur. Irregular fever with periods of apyrexia, the presence of œdema and enlargement of the spleen, anæmia and increase of mononuclear leucocytes, are common in both diseases. Polyadenitis and the presence of trypanosomes in the puncture material from the enlarged lymphatic glands, or in the cerebro-spinal fluid, or in the peripheral blood, confirm the diagnosis. It must, however, be noted that enlargement of the lymphatic glands in kala-azar, though not common in India, has also been observed in other affected areas.

In later stages kala-azar may be mistaken for *chronic malaria*, *tuberculosis*, *multilobular cirrhosis of the liver*, *Malta fever*, *Banti's disease*, and *generally diseases that are associated with low fever, anæmia without leucocytosis* and *chronic splenomegaly*. In *malarial cachexia*, as in kala-azar, the temperature may not be higher than 37·5° C. (99·5° F.) for many days. In most cases, however, the characteristic temperature curve can be made out, as well as the presence of malarial parasites in the blood and the characteristic action of quinine. It may be stated that an enlarged spleen which is soft, even after an illness of many months' duration, and which has become enlarged rather quickly, and has been steadily enlarging during apyrexial periods, or without any febrile exacerbations, is an indication of kala-azar rather than malaria. Although the spleen may become hard in chronic cases of kala-azar, yet some of the hardest and biggest spleens are found in malaria. If the spleen reaches up to the navel, during a course of illness of not more than two or three months, the diagnosis is more in favour of kala-azar than malaria.

The borderland cases between kala-azar and malaria present the greatest difficulty in diagnosis. Not infrequently one may meet with cases with the following blood-picture: Red blood-cells, 2,500,000; white blood-cells, 3,000; haemoglobin, 30 per cent. Such a blood-picture may be present in both malaria and kala-azar. In such cases marked diminution in the number of the polymorphonuclear leucocytes is an indication of kala-azar. In these cases a spleen puncture should be made after administration of calcium chloride, and a culture should be made from the splenic juice and peripheral blood on N N N medium for flagellates, if there are facilities. The globulin tests should be made. Bleeding from the skin and mucous membranes, and disturbances of the alimentary canal are more common in kala-azar than in malaria, especially in the case of children.

Archibald has observed that *intestinal schistosomiasis* caused by *Schistosoma mansoni* may produce fever associated with splenomegaly. In such cases one should look for leucocytosis and examine the stools for the presence of the characteristic ova.

Ankylostomiasis may be associated with fever and sometimes, according to certain observers, enlargement of the spleen without leucocytosis. The presence of eosinophilia, the blotting-paper tongue, the presence of worms and their ova in the stools, should make the diagnosis easy. This disease may be a complicating factor in kala-azar, and in cases where there is extreme anæmia, its presence should be suspected.

Histoplasmosis, a rare disease described by Darling, closely resembles kala-azar of adults in its symptoms, which include irregular fever, enlargement of the spleen and liver, severe anæmia and leucopenia. The causative organism, which was considered by Darling to be a protozoon, has been shown by Rocha-Lima to be a yeast-like body belonging to the cryptococci.

Toxoplasmosis, another rare disease first described by Castellani, may resemble kala-azar in its symptoms. The disease was supposed by Castellani to be due to a parasite described as *Toxoplasma pyrogenes*. Wenyon holds that no such parasite exists. Probably the cases described by Castellani were cases of kala-azar.

Malignant diseases of the abdomen may be associated with chronic irregular fever, and if not carefully examined may be mistaken for kala-azar.

The possibility of the occurrence of *Banti's disease* in the endemic areas of kala-azar should be borne in mind. There is a record of a museum specimen of a spleen removed for Banti's disease, in which abundant kala-azar parasites were subsequently found. The common symptoms of the two diseases are gradually increasing enlargement of the spleen, anæmia, leucopenia (average leucocyte count 3,500) with hæmorrhages, a tendency towards development of ascites, a progressive cachexia and a gradual down-hill course. In Banti's disease, however, one observes absence of the characteristic double rise of temperature. The history of frequent hæmatemesis, the extraordinary chronic course of the disease lasting for ten or twelve years with an enlargement of the spleen, which may not cause any discomfort, the development of jaundice with ascites in the late stage of the disease, and the absence of Leishman-Donovan bodies in the spleen help in its diagnosis. It is very important that a careful differential diagnosis be made in suspected cases in endemic areas of kala-azar, as early removal of the spleen in Banti's disease may lead to complete recovery.

Pernicious anæmia may sometimes be associated with leucopenia and enlargement of the spleen, but the high colour index, the large number of nucleated red corpuscles, and the clinical course should enable one to distinguish the disease from kala-azar.

Hodgkin's disease may be confused with kala-azar. Some of its symptoms are, low pyrexia with enlargement of the spleen, severe anæmia of the chlorotic type in late stages, some relative increase of the lymphocytes with proportionate diminution of the polymorphonuclear leucocytes. Sometimes there may be recurring attacks of pyrexia, and there is enlargement of the abdominal glands only. In such cases the disease may be possibly mistaken for kala-azar. Generally speaking, only a moderate enlargement of the spleen with marked enlargement of the lymphatic glands is a characteristic feature of the disease. Presence of *Leishmania* should establish the diagnosis.

Multilobular cirrhosis of the liver may be mistaken for kala-azar cirrhosis. In both there is ascites, hardening of the liver, and there may be leucopenia. Generally speaking, there is no history of fever in multilobular cirrhosis. The enlargement of the spleen is much greater in kala-azar. In doubtful cases spleen puncture should be performed with very great care.

Subacute ulcerative endocarditis with fever and enlargement of the spleen is diagnosed from kala-azar by the presence of cardiac signs and symptoms, and absence of leucopenia. Leucocytosis and optic neuritis, if present, are much against a diagnosis of kala-azar. Cases in which there is absence of cardiac symptoms and signs may present difficulty in diagnosis. In all suspected cases a blood-culture should be made for infecting micro-organisms of ulcerative endocarditis on the one hand and for flagellates on the other. Positive results will help in the diagnosis. Negative results are not so diagnostic, especially in the case of ulcerative endocarditis.

Leukæmias are easily differentiated from kala-azar by the presence of high leucocytosis.

TABLES OF DIFFERENTIAL DIAGNOSIS.

TABLE I.—KALA-AZAR AND MALARIA.

	(1) Fever	(2) Blood examination	(3) Spleen	(4) Liver	(5) Spleen and liver puncture
Kala-azar in early stages	Characteristic double rise of temperature, generally remittent, sometimes intermittent with single or double intermission during 24 hours. Long-continued double rises during 24 hours are suggestive of kala-azar. Rigors not so common	L.-D. bodies, if found in the peripheral blood, diagnostic. A progressive leucopenia more marked than in malaria. Ratio of W.B.C. to R.B.C. may be 1 to 1,500, or less. Culture of peripheral blood on NNN medium shows leishmania. Relative increase of lymphocytes or rarely large mononuclears	Rapid marked enlargement of the spleen	Frequently enlarged	L.-D. bodies frequently found in the smear from spleen puncture, may be found in the smear from liver puncture
Acute malarial fever	Characteristic curve of one or other varieties of malarial fever. In double tertian or malignant tertian infection, fever may be quotidian. In multiple or malignant tertian infection, it may be remittent. In the case of the latter, double rises of temperature, if present, are not long continued. The tendency to sudden high rises of temperature is specially to be expected in malaria. Rigors more common	One or other forms of malarial parasites present, if blood is examined before quinine has been administered or after quinine has been stopped. Ratio of W.B.C. to R.B.C. more than 1 to 1,000. Rarely, it may be less. Pigmented leucocytes present	Spleen less quickly enlarged than in kala-azar	Not much enlarged early in the disease, though hepatic tenderness is not infrequent	Malarial parasites present in the smears

	(6) Jaundice	(7) Appetite	(8) Early œdema	(9) Quinine test	(10) Globulin test	(11) Aldehyde test
Kala-azar in early stages	Rare	Good in many cases	May sometimes be present	No effect	Frequently positive	May be positive
Acute malarial fever	Slight icteroid tint is not uncommon	Generally bad	Not present	Quinine stops the fever generally after oral administration. Rarely quinine may have to be given intramuscularly or intravenously before fever stops	Almost always negative	Almost always negative

In addition to many of the diseases mentioned above, infantile kala-azar has to be differentiated from diseases of children associated with enlargement of the spleen. One of these is *infantile cirrhosis of the liver*. Its features are enlargement of the spleen and liver, irregular rises of temperature, especially in the early morning, early appearance of ascites and marked jaundice, leucocytosis and absence of *Leishmania* in the spleen. On the other hand, a case in a child with slight ascites and enlarged spleen and liver, with a history of prolonged fever without much change in the leucocyte count, may be mistaken either for kala-azar or infantile cirrhosis of the liver.

TABLE II.—KALA-AZAR AND TYPHOID FEVER.

	(1) Fever	(2) Blood examination	(3) Spleen	(4) Liver	(5) Lungs
Kala-azar in early stages	See Table I.	See Table I. Widal reaction negative	See Table I.	See Table I.	Bronchitis generally absent
Typhoid fever	Step-ladder-like rise of temperature and a characteristic course. Rogers lays stress upon high continued fever specially in the second week. He regards this as diagnostic of this disease. Rigors not common	Widal reaction—positive after 10 days' illness. Leucopenia not so marked, except in some cases towards late convalescence. Blood-culture may show typhoid bacillus even after 2 or 3 days' illness	Enlarged but not so rapidly marked enlargement	Only slight enlargement may be present	Bronchitis frequently present. Also broncho-pneumonia may be present

	(6) Early œdema	(7) Constitutional and general symptoms	(8) Abdominal symptoms	(9) Diazo reaction	(10) Pulse	(11) Aldehyde test and globulin test
Kala-azar in early stages	See Table I.	Generally not present in the ordinary type of the disease	Generally not marked in the ordinary type of the disease. Hæmorrhage from the bowels rarely present. Tongue generally clean	Absent	Frequently quick from the very beginning and may be out of proportion to the temperature	See Table I.
Typhoid fever	Not present unless complicated by nephritis	Generally present in the ordinary moderately severe cases	Fairly marked in the ordinary cases. Hæmorrhage from the bowels more common. Tongue dry and cracked in severe cases. Sometimes coated or furred in the centre with red tip and edges	Frequently present	Slow in comparison with the temperature, frequently dicrotic. Atropine test (Marris) positive, specially in the second week	Almost always negative

TABLE III.—KALA-AZAR AND PARATYPHOID FEVER.

	Fever	Blood examination	Spleen	Early œdema	Pulse	Aldehyde test and globulin test
Kala-azar in early stages	See Table I.	See Table I. Widal reaction for paratyphoid bacillus (A or B) negative	See Table I.	See Table I.	See Table II.	See Table I.
Paratyphoid fever	Course of the fever generally a fortnight	Widal reaction for paratyphoid bacillus (A or B) positive after the fever has lasted for 10 days or more. No marked leucopenia. Blood-culture may show the growth of paratyphoid bacillus (A or B)	Enlarged but not so rapid enlargement	Not present	May be slow in comparison with the temperature. Atropine test (Marris) positive, specially in the second week	Almost always negative

Rickets in children may sometimes be associated with much enlargement of the spleen and the liver and, in the absence of definite signs of the disease, might be mistaken for kala-azar. Generally speaking, the characteristic changes of the bones in the disease, leucocytosis, and absence of disproportion between the polymorphonuclears and lymphocytes, are of diagnostic value.

Splenomegalic cirrhosis of the liver occurs in children or young people. It is characterized by marked enlargement of the spleen, slight enlargement of the liver, anæmia without leucocytosis, hæmatemesis, jaundice, clubbing of the fingers, and stunted growth. A careful study of the symptoms will help in differentiation of the two diseases.

Infantile splenomegaly without *Leishmania*, as observed by Nicolle and others, occurs in Mediterranean countries and in the Sudan. It is characterized by pyrexia and enlargement of the spleen and the liver, and the absence of parasites.

Hanot's cirrhosis (also called hypertrophic biliary cirrhosis) may be mistaken for kala-azar. It occurs frequently in children and lasts for several years. It is characterized by enlargement of the spleen and the liver, sudden rises in temperature, attacks of pain in the hepatic region, stunted growth, clubbing of the fingers, the late appearance of ascites and a tendency to hæmorrhage. The history of marked jaundice for years, without any marked illness, and presence of leucocytosis and absence of *Leishmania*, are indications that the disease is not leishmaniasis.

Congenital syphilis may be associated with enlargement of the spleen. The absence of *Leishmania*, the family history, the consideration of other symptoms and the Wassermann reaction, will differentiate the disease from leishmaniasis.

Infantile pseudoleukæmia (von Jaksch's disease) may present difficulty in diagnosis in an endemic area of kala-azar. It affects children under two years of age. The symptoms are hæmorrhage from the mucous membranes, periodic attacks of fever, enlargement of the spleen and the liver, and ascites. The blood-picture shows marked anæmia with low colour index, generally a

TABLE IV.—KALA-AZAR AND GENERAL TUBERCULOSIS.

	(1) Fever	(2) Blood examination	(3) Spleen	(4) Lungs	(5) Constitutional symptoms
Kala-azar in early stages	See Table I.	See Table I.	See Table I.	Bronchitis generally absent	See Table II.
Acute general tuberculosis	Irregular fever. There may be an inverse type of temperature	There may be leucocytosis, though leucopenia is more common, rarely tubercle bacilli may be cultivated from the blood	May be enlarged, but is neither so early nor so markedly enlarged as in kala-azar. In children sometimes may be much enlarged	There may be diffuse bronchitis. Respiration increased in frequency, specially in the early stage. There may be signs of bronchopneumonia. Cyanosis may be present. Sputum may show tubercle bacilli	Torpor and dullness gradually deepening into coma. There may be active delirium

	(6) Diazo reaction and tuberculin test	(7) Lumbar puncture	(8) Tongue	(9) Jaundice	(10) Ophthalmoscopic examination	(11) Aldehyde test and globulin test
Kala-azar in early stages	Absent	Negative	Generally not dry	Rarely present	Nothing abnormal	See Table I.
Acute general tuberculosis	May be positive	There may be tubercle bacilli	More commonly dry	May be present	Choroidal tubercles present	Absent

TABLE V.—KALA-AZAR AND *Bacillus coli* INFECTION.

	Fever	Blood examination	Early œdema	Spleen and liver	Urine
Kala-azar in early stages	See Table I.	See Table I.	May be present	Generally enlarged	Nothing peculiar
Bacillus coli infection	Irregular temperature curve. Frequent severe rigors with rapid high rise of temperature, sometimes more than once in 24 hours. Sometimes the temperature resembles that of typhoid	Well-marked leucocytosis	Absent	Not enlarged	Acid, frequently opalescent. Motile rod-shaped bacteria present in the urine. Culture shows pure growth of *Bacillus coli*. May contain pus. Symptoms referable to the urinary system generally present in long-standing cases

TABLE VI.—KALA-AZAR AND CHRONIC MALARIA.

	(1) Fever	(2) Blood examination	(3) Spleen	(4) Liver	(5) Spleen and liver puncture
Kala-azar in its chronic stage	May be low continued or intermittent rising to 99° or 100°. There may still be tendency towards double rise. Rigors—rare	L.-D. bodies if found are diagnostic. A progressive leucopenia. Leucocyte count frequently about 2,500, may be 800. Ratio of W.B.C. to R.B.C. may be 1 to 1,500, or even less. Culture of peripheral blood shows leishmania. Increase of lymphocytes more common	Generally much enlarged. A moderately enlarged spleen with œdema which cannot be accounted for by any heart or kidney disease is suspicious of kala-azar	Enlarged and sometimes much enlarged. Cirrhosis of the liver with its attendant symptoms may be present in some cases. It is generally firm and hard. Enlargement of the liver is more marked in kala-azar than in malaria	L.-D. bodies frequently found on spleen puncture. They may also be found on liver puncture
Chronic malaria	Characteristic curve of one or other varieties of malarial fever. In some cases the temperature may be low for days not going above 99·5°. There is, however, a tendency towards sudden high rises of temperature even in these cases from time to time. In some cases the fever may be of an irregular type. Rigors — more common	One or other forms of malarial parasites present, if blood is examined before quinine has been administered or after quinine has been stopped. Generally speaking, the ratio of W.B.C. to R.B.C. is greater than 1 to 1,000; sometimes though it may be less. Leucopenia generally not so marked as in kala-azar. Pigmented leucocytes present. Increase of large mononuclears more common	Spleen may be much enlarged	Liver may be enlarged, but generally there is no cirrhosis of the liver in malaria	Malarial parasites may be found

	(6) Appetite	(7) Quinine test	(8) Skin and general appearance	(9) Urine	(10) Aldehyde test and globulin tests
Kala-azar in its chronic stage	Good in many cases	No effect	Peculiar, shining stretched appearance of skin. Wasting—marked	Slight albuminuria with urobilinuria is diagnostic (Knowles)	Frequently positive
Chronic malaria	Generally marked loss of appetite	Quinine generally stops fever after oral administration for a sufficient length of time. Rarely, quinine may have to be given intramuscularly or intravenously before fever stops	Peculiar pale yellow tinge in skin. Wasting less marked. Marked anæmia without much wasting is suggestive of malaria	Albuminuria not common	Frequently negative

TABLE VII.—KALA-AZAR AND MALTA FEVER.

	Fever	Blood examination	Spleen and liver	Joint symptoms	Serum test	Culture from blood and urine
Kala-azar in its chronic stage	See Table VI	See Table VI	See Table VI	Not present	Serum does not agglutinate emulsion of *M. melitensis* in proper dilution	No growth of *M. melitensis*
Malta fever	There are waves of intermission and of more or less pyrexia of variable length	There may be slight leucocytosis	May be enlarged but generally not so much as in kala-azar	An acute or subacute effusion may take place suddenly into one or more joints, without any apparent reason	Serum agglutinates emulsion of *M. melitensis* in proper dilution	Growth of *M. melitensis* from blood and urine

TABLE VIII.—INFANTILE KALA-AZAR AND INFANTILE BILIARY CIRRHOSIS.

	Fever	Blood examination	Spleen and liver	Spleen and liver puncture	Jaundice	Aldehyde test and globulin tests
Kala-azar in its chronic stage	See Table VI	See Table VI	Generally spleen is enlarged out of proportion to enlargement of liver. Liver not so hard and enlarged in the beginning as in infantile biliary cirrhosis	See Table VI	Rare	Positive
Infantile biliary cirrhosis	Low fever generally rising towards early morning	Leucocytosis frequently marked	Liver enlarged out of proportion to enlargement of the spleen, which may not be enlarged in some cases. Liver enlarged, hard and irregular from the beginning and ascites may be early; it is frequently marked in advanced cases	Negative	Very marked and early	Negative

leucocytosis, especially lymphocytosis, nucleated red cells, megaloblasts and cells containing intensely polychromatophilic cytoplasm.

Still's disease is a disease of children, associated with general destructive affections of the joints, enlargement of the spleen and lymphatic glands, emaciation, anæmia, and a tendency to pigmentation. The disease in its developed stage is unmistakable, owing to the condition of the joints.

Gaucher's type of splenomegaly is a familial disease which starts early

in life and runs a chronic course. It is characterized by enlargement of the spleen followed by that of the liver, but there is no jaundice, ascites or discoloration of the skin. When anæmia and leucopenia are marked, it may be mistaken for kala-azar. The more chronic course of the disease and the absence of *Leishmania* should confirm the diagnosis.

Pseudo-kala-azars, as defined by Castellani, are "febrile or afebrile diseases which resemble kala-azar, in that they are associated with splenomegaly, anæmia and often emaciation." They are differentiated from kala-azar by absence of *Leishmania*. Rogers holds that malaria and kala-azar account for all the common cases of splenomegaly in Bengal. The author has met with a large number of cases of splenomegaly without *Leishmania* or malarial parasites. The question arises as to whether some of these are sequelæ of kala-azar in which *Leishmania* have disappeared from the spleen or exist in some undiscovered phase. The possible existence of another undiscovered causal micro-organism is also unproven.

Cases of *obscure anæmia with œdema*, without enlargement of the spleen, in an endemic area, should have their blood properly examined, as such cases are sometimes due to kala-azar.

The various causes of enlargement of the spleen are quoted below from French's *Index of Differential Diagnosis*:—

"I. Chronic enlargement of the spleen.

"(*a*) *Very Great Enlargement.*—Spleno-medullary leukæmia, lymphatic leukæmia, mixed leukæmia, chronic malaria, kala-azar, splenomegalic polycythæmia, splenomegalic cirrhosis, splenic anæmia, pseudo-leukæmia infantum, Gaucher's disease, Still's disease, familial acholuric jaundice, Egyptian splenomegaly.

"(*b*) *Moderate Enlargement.*—All conditions mentioned in group (*a*) will at some stage exhibit a spleen that has not yet become enormous; and besides these, chronic and moderate enlargement of the spleen may be exhibited in cases of pernicious anæmia, rickets, congenital syphilis, Hodgkin's disease, cirrhosis of the liver, lardaceous disease, thrombosis of the portal vein, pressure on the portal vein by enlarged lymphatic glands or by adjacent tumour of the gall-bladder, liver, pancreas, stomach, &c.

"II. Acute enlargement of the spleen, the enlargement as a rule being slight.

"(*a*) Acute infective fevers.—Specially typhoid fever, paratyphoid fever, relapsing fever, typhus fever, malaria, Malta fever, erysipelas, septicæmia. Less often in pneumonia, diphtheria, scarlet fever, small-pox, rheumatic fever, influenza, general tuberculosis.

"(*b*) Embolism, specially in cases of fungating endocarditis.

"(*c*) Injury.

"(*d*) Strangulation by twisting of the pedicle."

To the above list of acute enlargements, kala-azar has to be included.

It will be noted that no mention is made of abscess, gumma, or tumours of the spleen, as they are very rare, nor has mention been made of causes of backward pressure, such as chronic valvular heart diseases with failing compensation. The latter rarely produce splenic enlargement, except in children.

CHAPTER XIII.

PATHOLOGY.

SHORTT has observed that in experimentally infected monkeys there sometimes occurs an acute and fatal type of the disease. Generally the disease is chronic in these animals.

The liver is slightly enlarged and fatty, and parasites are present in the endothelium of the finest capillaries, blood sinuses, and also in the Küpffer cells. The spleen is considerably enlarged, the Malpighian corpuscles are prominent, there is hyperplasia of the endothelial cells, and their distension by the parasites may sometimes be so great as to appear to occlude the lumen of the sinuses.

Parasites are also present in the spleen inside large macrophages, polymorphonuclear and occasionally eosinophile leucocytes. The bone-marrow is dark in colour, because the yellow marrow is replaced by red marrow. The parasites are found in the same cells as in the spleen and liver. The bone-marrow contains the largest number of parasites. There is a preponderance of giant cells in this tissue. In the testes, parasites were found by Shortt in largest numbers in the interstitial cells of Leydig, but they were not found in the endothelium of large vessels. In the lungs and kidneys parasites have been found in the endothelial cells.

In experimentally infected mice the spleen is always enlarged, and parasites are found in the spleen, liver, bone-marrow, and the testes. In experimentally infected dogs, the splenic enlargement is not so marked as in monkeys. The parasites have been found in the spleen, bone-marrow, liver, lungs, mesenteric glands and kidneys.

Meleney has studied the changes present in artificially infected hamsters. In these a specific tissue reaction occurs in the form of endothelial proliferation, forming masses of "clasmatocytes" heavily parasitized. In definite patches of clasmatocyte tissue, the most striking thing is the slight degree to which the cells are parasitised. Most of them contain a few or moderate number of parasites, but some cells, particularly in the centres of the cell masses, contain none. At the periphery some may be crowded with parasites. Wherever fat appears, single clasmatocytes, heavily parasitized, are scattered through it. The chief organs affected are the liver, spleen, lymph nodes and bone-marrow. He considers that in advanced infections the parasites also attack the parenchymatous cells of the liver and adrenal cortex, extensive degeneration of the liver occurs, and infected "clasmatocytes" appear in practically every organ and tissue, the stroma of the intestinal mucosa being

the site of massive accumulation of those cells. The most favourable sites for the localization of the parasitized clasmatocytes are the regions where loose areolar tissue is associated with good vascularity.

In one case Wenyon found flagellate forms in the spleen of a dog infected with leishmania from a case of Indian kala-azar. There is no evidence that the leishmania exist in flagellate forms in the infected host, with this single exception. In another case, Maitra found, in a film made from the peripheral blood of a case clinically diagnosed as one of kala-azar, flagellates of the leptomonas type. The slide containing the flagellates was subsequently shown to the author. As the flagellates were found in only one of the films

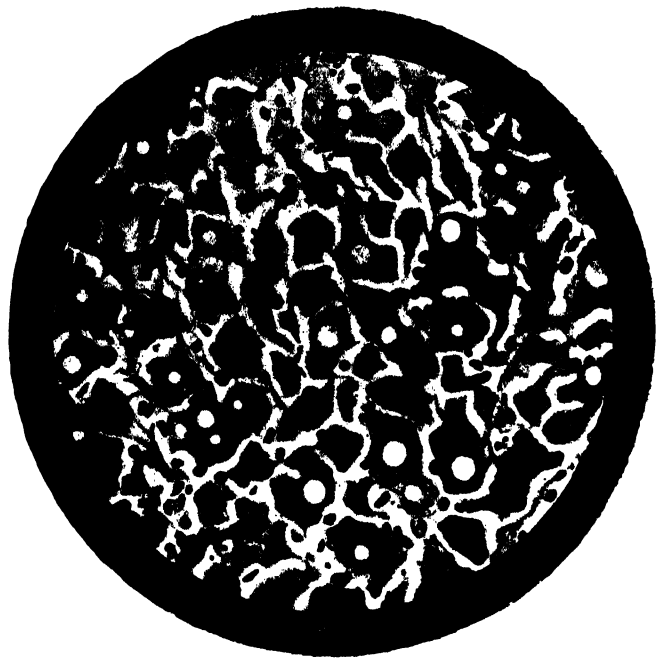


FIG. 22. Fatty liver in kala-azar. (Original.)

made at one and the same time, the possibility of the film having been contaminated cannot be excluded.

The process of infection in kala-azar probably takes place in the following way (Christophers): The pathogenic micro-organisms are taken up by or invade the endothelial cells in the visceral or other capillaries, e.g., of granulation tissue. The endothelial cells (macrophages) increase in size and become more and more distended, due to the proliferation of the parasites within their cytoplasm, and subsequently become converted into mere bags filled with large numbers of the parasites. Eventually such cells rupture and the parasites are set free, to be taken up again by other endothelial cells, polymorphonuclear or mononuclear leucocytes, and carried to the peripheral

circulation. It was Christophers who first showed that the disease appeared to be essentially an infection of the vascular endothelium and resembled chronic septicæmia. The author holds that probably it is principally of the nature of an infection of the reticulo-endothelial system distributed in the different parts of the body, and especially of the spleen, liver and bone-marrow. The enlarged parasite-laden cells found in these organs in kala-azar mostly correspond to those of the reticulo-endothelial system which swell up and enlarge in size by stimulus of the presence of dye-stuffs in the process of vital staining. The clasmatocytes probably belong to the reticulo-endothelial system. It may be stated that apart from what has been considered to be a specific tissue reaction in the form of appearance of the clasmatocyte tissue, one agrees with Shortt "that Christophers' original description, in 1904, of the morbid histology of the disease in man has left little for succeeding workers to describe or amend." In this connection it may be noted Shortt considers that so close is the connection between the endothelium of capillaries, stroma cells, and macrophages, that one cannot help considering the probability that the original stroma cells produce either of the other two types, given the appropriate stimulus.

Skin.—Christophers found the parasites in the endothelial lining of very fine cutaneous capillaries in the papular eruptions of the skin in advanced cases, about the thighs, Scarpa's triangle and the scrotum. He also found them in ulcers and in minute nodules of the skin. On section, the nodules showed signs of inflammation, widening of the vessels and small round-celled infiltration. Recently the author has found a large number of *Leishmania* in the skin in cases of Dermal Leishmanoid, a disease which sometimes develops in cases of kala-azar which have been cured by antimony (see Dermal Leishmanoid). According to Laveran, no cutaneous ulceration with *Leishmania* has been found in infantile kala-azar. These may, however, occasionally be present in ulcers of the skin in infantile kala-azar. These cutaneous ulcers in infantile kala-azar, in the first stage of their development, appear as a number of irritating papules. Later on they change into painless nodules, in the centre of which is a small opening whence a fluid flows. The fluid dries into a crust. A number of nodules sometimes form a group, from which first a blister and then an ulcer develops. This has smooth edges and a granular surface. On making a section the position of the ulcer, near to the sweat glands and hair follicles, is revealed. In the superficial layers an infiltration consisting of cells resembling plasma cells is to be seen. In addition to these, other cells with granular protoplasm are present. Signs of *capillaritis obliterans* may be seen in various positions. (For other peripheral lesions see Addendum.)

Fat Tissue.—In infected hamsters, wherever fat appears, single clasmatocytes, heavily parasitized, are scattered through it (Melency).

Lymphatic Glands.—Leishmania were found by Christophers in lymphatic glands, especially in ulcers over the skin near to them. Cochran found them in smears from excised lymphatic glands.

The mesenteric glands are swollen and soft. Jemma and di Cristina

state that the sinuses may be widened, the endothelium may be peeled off and may contain *Leishmania*. The fibres of the reticulum are thickened. The endothelial cells may atrophy and there may be signs of necrosis (Dionisi). There may be preponderance of parasite-laden cells in the follicular spaces.

Lymph Nodes.—Parasites may be found in the loose reticulum, and the degree of parasitization of any one group of clasmatocyte cells seems to be in inverse proportion to the number of cells in the group.

Intestine.—During the course of the disease catarrhal changes and sometimes ulcers are found in the lower part of the ileum. These are of different sizes, their border is irregular and they have a reddish-grey base. Inflammation of the intestinal mucous membrane frequently occurs, and if it is of long duration it may lead to attenuation of the intestine throughout its length. Microscopic examination shows that the ulcer slowly destroys the superficial layers of the mucous membrane, penetrates deeply and may reach the muscular coat. Granulation is seen in the surrounding tissue.

Mackie has found bodies resembling *Leishmania* in the smears of the stools of patients suffering from Indian kala-azar, in whom dysenteric symptoms had been induced by the administration of croton oil. *Leishmania* have also been found in the endothelial cells of fine capillaries in ulcers of the large intestine. Intestinal ulcers are more common in Indian kala-azar, in which multiple ulceration may be found in the large intestines which may extend deeply into the muscular coat. Christophers found *Leishmania* as intracellular parasites in the endothelial cells lining the blood-vessels in the granulation tissue of these ulcers. They may also be found inside the macrophages.

Jemma and di Cristina, in their observation of cases of infantile kala-azar, noted the constant occurrence of enterocolitis, and the presence of circular ulcers with elevated edges in the large intestine.

Perry has observed that the jejunum may appear slightly thickened, without any ulceration. Microscopically the following histological changes were noted by him :—

"Each villus was transformed into a swollen, distorted and polypoid body, connected with the submucous tissue by a constricted stalk, formed of a few fibres of connective tissue. The columnar epithelium covering the villi had disappeared and the basement membrane furnished a delicate limiting sheath for each little swelling. The internal structure of the villi was completely altered, owing to an intense proliferation of the endothelial cells lining the lymph channels. This proliferation of endothelium, although marked in the base of the villi, became more pronounced towards the centre and extremities of these structures, and the enlargement and distortion were caused by these tightly-packed accumulations of hypertrophied cells.

"In the greater number of villi the basement membrane was intact, but in many instances it had ruptured from over-distension and liberated the enclosed endothelial cells.

"The distribution of Leishman-Donovan bodies in the intestine was very striking. They could be demonstrated in scanty numbers in the submucous coat ; in that position they occurred in endothelial cells evidently derived

from vascular endothelium. They were present in larger numbers, in the same intracellular situation, in the base of the villi. In the centre of the villi they had undergone rapid multiplication, and they were present in enormous numbers in the endothelial cells, distending the extremities of these structures. In many of the villi numbers of endothelial cells had broken down, and the parasites were lying free in the villus mixed with the debris of necrotic cells." Perry emphasizes the possibility of the escape of viable parasites by the intestines.

In the hamster, Meleney found similar changes in the gastro-intestinal tract, the submucosa being invaded by parasitized macrophages, especially near to Peyer's glands and solitary lymphatic nodules, the epithelium being intact. Similarly in man, he found that where lymph follicles occurred, parasitized clasmatocytes were numerous in the neighbouring submucosa, the ileum, cæcum and colon being less infected than the stomach and jejunum.

Banerjee has found the parasite in the ulcers of the stomach of a case of kala-azar.

Dionisi found *Leishmania* in the follicles of the intestines in cases of infantile kala-azar. In one case of infantile kala-azar Critien found *Leishmania* in the stools.

Pancreas.—There are no visible changes in the pancreas. Pianese found that the islets of Langerhans underwent hypertrophy and hyperplasia, and also discovered *Leishmania* in the large mononuclear leucocytes.

Lungs.—*Leishmania* are found in the large mononuclear and polymorphonuclear leucocytes. Parasitizing cells of the vascular endothelium and clasmatocytes may be present.

Mammary Glands.—The mammary glands are atrophied in a large number of cases (Dionisi).

Testes.—*Leishmania* may be found in the capillaries. As already stated, in experimental animals, Shortt found them inside the cells of Leydig. Meleney found them inside clasmatocytes in man.

Kidneys.—The kidneys sometimes show no changes. There may be congestion, hæmorrhages or signs of nephritis in the kidneys. Parasites have been found in the renal vessels, glomerular capillaries, and recently Shortt has cultured the parasites from the urine of persons suffering from Indian kala-azar. According to Archibald, the parasites have never been found in the cells of the secreting tubules. They have been found inside cells of the reticulo-endothelial system in experimental animals.

Adrenal Glands.—*Leishmania* may be present in the endothelium of the vessels in the cortex and medulla. Jemma and di Cristina found a glandular cell of the cortex invaded by *Leishmania*. In experimental animals, the parenchyma of the cortex may be infected.

Heart and Blood-vessels.—In the large veins parasites have been found inside endothelial cells. In the heart they have been found inside polymorphonuclear leucocytes. In small and medium-sized vessels, fatty degeneration of the intima is found. Many investigators have found the parasites in the peripheral blood. In 1909 Donovan stated that in 92 to 93

per cent. of his cases he found the parasites both in mononuclear and polynuclear white blood-corpuscles. Patton found the parasites in the peripheral blood of thirty-eight out of forty-five cases which he treated in the General Hospital at Madras. In the Sudan, Thomson and Marshall found them in thirteen out of fifteen cases, and Cannata, Rutelli and Vaglio found them in 93 to 95 per cent. of their cases in Italy. The *Leishmania* are seen in the large mononuclear cells, which Patton and Christophers consider to be endothelial cells which have broken loose. In addition they are also found inside polymorphonuclear leucocytes. Knowles examined a series of 73 cases, and on 33 occasions he found parasites in the blood, i.e., 45 per cent., while 10 per cent. only of 682 films, made from the blood of these cases, gave a positive result. He considers that if the blood films are properly made then the findings may be more frequently positive. Examination of thicker film smears gave much better results in his hands.

Some experiments in which suprarenin (adrenaline) injections were given in order to bring the parasites more into the peripheral circulation, were not successful (Mazzoni). Knowles, however, stated that he obtained positive results more frequently when the patient had been given a subcutaneous injection of one cubic centimetre of 1 in 1,000 adrenaline solution half-an-hour before the blood was examined. By making cultures from the blood on N N N media, as described in the Chapter dealing with diagnosis, very frequently positive results are obtained, and sometimes even as early as the fourth day.

As regards the number of parasites present in the peripheral blood at various stages of the disease, Patton is of opinion that they are present in greatest abundance in the last stages of the disease, and in those cases which are complicated by dysentery. Others, however, have observed the parasites in greatest numbers in the early stages, as well as during the earlier and recurrent attacks of dysentery and during fever. Apparently any condition which raises the leucocyte count of the peripheral blood (e.g., dysentery and broncho-pneumonia) increases the possibility of finding the parasites in the peripheral blood. According to Knowles, the parasites are easy to find when myelocytes are present.

For further changes in the blood-picture, see Chapter IX.

In infected hamsters, heavily infected clasmatocytes are found in the interstitial tissue throughout the myocardium and subendocardial tissue of the heart.

The pathology of dermal leishmanoid is described in Chapter IX.

Nervous System.—*Leishmania* have been found by Christophers in the polymorphonuclear cells, in petechiæ from the arachnoid of Indian kala-azar. There is no record of *Leishmania* being present in the cerebro-spinal fluid of Indian kala-azar. La Cava found them in the cerebro-spinal fluid of infantile kala-azar.

Muscles.—Visentini found parasites in the muscles, in one case which he investigated.

Spleen.—The spleen is often swollen and sometimes it is enlarged to an

enormous degree. In the pathological museum of the Calcutta Medical College, there is a specimen of a kala-azar spleen of an adult weighing nearly three kilogrammes. The author had a case in the Calcutta Campbell Hospital in which the spleen weighed six and a half kilogrammes.

Its external form does not change very much, although its contours become more rounded and the segmentation more marked. The surface sometimes appears opaque, because of the perisplenitic changes. Adhesions to adjacent organs may occur. In some places the follicles are visible, while in others they are not. The venous sinuses are dilated, and hæmorrhages may occur. In acute cases the capsule of the spleen is generally smooth and stretched, and does not adhere to the adjacent organs.

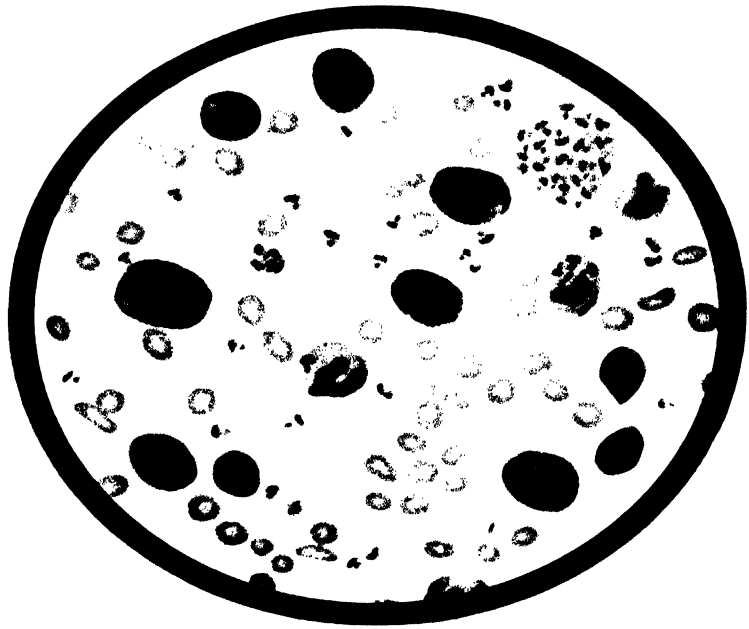
The consistency of the splenic parenchyma is variable. In acute cases the spleen pulp is soft and bulges out when the capsule is cut. It may rupture during exertion or through injury. In chronic cases the firmness of the spleen increases, the capsule is thick, and some observers consider that there is less danger of its rupturing. The author does not agree with the latter view. The spleen presents a solid appearance and keeps its form after having been removed. The substance is of a deep red colour and is granular in appearance. The trabeculæ are prominent. The spleen is firm to the touch, although it is friable. Anæmic infarcts may be present. It is stated by some that the violet-red colour of the splenic parenchyma is different from the slate colour of the malarial spleen.

Sometimes the Malpighian corpuscles are hypertrophied and sometimes they are atrophied and less prominent than normal. The splenic reticulum, which is scarcely apparent in subjects who succumb at an early period of the disease, may become thick, especially in the region of the follicular artery, in chronic cases. This is, to some extent, the cause of the splenic hypertrophy in these chronic cases. The follicular arteries also become thickened, their endothelium is swollen and permeated with parasites. Numerous *Leishmania* are also to be found in the enlarged follicular cells. Here and there the connective tissue loses its transparency. Uniform areas of connective tissue are formed in place of the degenerate follicular elements and cells of the reticulum (Dionisi). The sclerotic tissue of the follicles and pulp eventually develops from this.

In infantile kala-azar, di Cristina and Cannata found the capsule thickened, numerous endothelial elements swollen, and Malpighian corpuscles poor in lymphoid elements. The endothelium of the blood-vessels and vascular spaces contained *Leishmania*. Visentini found the reticulum excessively infiltrated with blood, and in some places where the distension of the reticulum reached its maximum, especially near to the capsule, there were isolated areas of degenerated parenchyma. The follicles were sometimes reduced in size, being represented by little heaps of lymphocytes shrivelled up round the arterioles.

Dionisi states that the follicles are either degenerated or atrophied. The cellular elements of the reticulum of the follicles are swollen and full of parasites. The meshes of the reticulum are thickened. The spleen pulp

FIG. 1.—Case of subacute kala-azar. Section of the spleen.
(1) Congestion of the pulp. (2) Dilatation of the reticulum and the sinuses with angiomatous spaces. (3) Slight increase in reticular tissue. (4) Slight fibrosis of the arteries. (Eye-piece No. 1, Objective Zeiss D.)



John Bale Sons & Danielsson, Ltd

FIG. 2.—Leishman-Donovan bodies in a smear of spleen (stained by Leishman's method).

degenerates or undergoes necrotic changes. According to him the *Leishmania* are found in largest number in the spleen, especially in the endothelial cells of the capillaries.

The following types of endothelial cells containing the parasites are described by Christophers (see also Addendum) :—

(1) Slightly modified endothelial cells, whose protoplasm sometimes tends to become proliferated in the form of buds.

(2) Round cells with extensive protoplasm and a round or oval nucleus. These cells sometimes resemble small mononuclear leucocytes.

(3) Very large cells lying inside capillaries. They may be extended along the capillaries or their cytoplasm may be massed together.

(4) Extremely large cells. The cytoplasm of many of these cells is reduced to a mere pellicle containing a mass of parasites. These cells appear to be on the point of rupture.

Besides these cells the parasites may be found inside the large mononuclear and polymorphonuclear leucocytes in the spleen (Plate VI, fig. 2).

Some of the parasites derived from spleen puncture appear free, while others are found inside the large endothelial cells (Plate VI, fig. 2). In some cases these endothelial cells break up into small globular masses containing a number of these parasites. These masses sometimes resemble red blood-corpuscles, and may give the impression that the *Leishmania* are contained in red blood-corpuscles.

According to Nicolle, the large mononuclear cells derived from the endothelium of the capillaries contain the parasites exclusively in the spleen.

In many cases of Indian kala-azar the author has observed the following changes in the spleen :—

The spleen is frequently congested, and in acute cases markedly so. In acute and subacute cases the venous sinuses may be so dilated that angiomatous spaces are formed. The spleen may be overfilled with blood and hæmorrhages may occur in its tissue (Plate VI, fig. 1). On section of the spleen, a multiplication of the macrophages, loaded with *Leishmania*, is observed. In chronic cases, aggregates of pigment sometimes may be found in the connective tissue of the trabeculæ. Pigmentation of the spleen is not, however, a constant sign, and may be absent in many cases. There is an increase of fibrous tissue, and spaces containing large mononuclear cells full of *Leishmania* are seen (Plate VII, figs. 1 and 2). Degenerative areas may occur in the pulp, resembling what have been described by Thayer and others in chronic malaria. Whether these changes are indicative of old malarial infection at an earlier period, or due to kala-azar itself, is difficult to state. In some cases there is a hyaline degeneration of the small arteries in scattered areas. In other cases there is a thickening of the outer coat of the arteries with small round-celled infiltration. This thickening may sometimes be very extensive, and may be associated with marked formation of fibrous tissue in the spleen. The thickened arteries are seen running along these thickened fibrous trabeculæ. Sometimes the arteries appear to be obliterated. Fibrous nodules may sometimes be seen on section of the spleen. These appear, in

some cases, to be situated on the sites of the obliterated arteries. Sometimes pigmented nodules can be seen here and there in the spleen, generally in the place where the atrophied Malpighian corpuscles were present. In some cases there is general infiltration of the spleen with melanin.

Infarcts may be present in the spleen. There may be perisplenitis with thickening of the capsule in chronic cases.

Some of the changes described above are similar to those that have been observed in cases of chronic malaria, and may suggest the possibility of a previous malarial infection.

Generally speaking, the consistency of the spleen is hard, even in early stages of the disease. In this stage the hardness is due mostly to the organ being packed with *Leishmania*-laden cells, and there is little or no development of fibrous tissue, and the spleen may regain its normal size under proper treatment.

In chronic cases the hardness is due partly to the presence of leishmania-laden cells and partly to fibrous tissue development. In many of these cases the former preponderates over the latter, and the organ regains its normal size under proper treatment as in early cases. In those cases in which there is excessive development of fibrous tissue, the organ does not regain its normal size under proper treatment. Such cases are rare, and a very hard spleen which clinically gives an impression of excessive development of fibrous tissue may be due mostly to the presence of *Leishmania*-laden cells, as can be seen post mortem. Meleney found in a case in man that the entire tissue was composed of a reticulum containing "clasmatocyte tissue."

The enlargement of the spleen is due partly to the above factors and partly to the dilated venous sinuses and congestion of the organ. In those cases in which the spleen is soft, the dilated venous sinuses predominate over the fibrous tissue formation. In certain cases, the enlargement of the organ is partly due to infarcts, which also influence the consistency of the spleen.

In experimental animals, Shortt found the Malpighian corpuscles distinct, and there was a hyperplasia of the endothelium affecting the lining cells of the capillaries and blood-sinuses. The cells of the endothelial tissue may in part be so distended with parasites as to occlude the lumen of the sinuses. Large macrophages, containing parasites and probably of the same origin, may be seen.

In the hamsters Meleney found, in chronic infection, huge heavily infected clasmatocytes practically occupying the splenic pulp and partly replacing the lymph follicles.

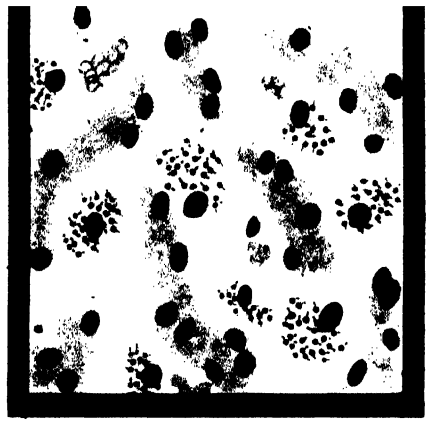
Liver.—The liver is often enlarged, although not to the same extent as the spleen. It is only in very rare cases that the liver appears more swollen than the spleen. Greyish-white spots, indicating the presence of perihepatitis, may be seen on the surface of the liver. The edges are regular and well-defined. The solidity of the organ is somewhat increased. The cut surface is reddish-yellow and has the appearance of nutmeg liver (Christophers). The lobulation is not recognizable. Microscopic examina-

PLATE VII.

Case of Chronic Kala-azar.

Fig. 1.—Section through a Malpighian body of the spleen. Fibrosis of the splenic tissue and obliteration of the central artery.

Fig. 2.—Marked fibrosis of the spleen with lacunæ in which are situated the L.D.-containing cells. (Eye-piece No. 1, Oil immersion $\frac{1}{12}$.)



John Bale, Sons & Danielsson, Ltd

Fig. 3.—Section of the liver. L.D. bodies in the Küpffer stellate cells (endothelial cells). A few red blood-corpuscles are seen in the capillaries. (Eye-piece No. 1, Oil immersion $\frac{1}{12}$.)

tion of the liver from advanced cases shows widening of the interlobular capillaries, and an increase in the amount of connective tissue present. The liver cells appear to be damaged in a varying degree. In addition to atrophy and disintegration of the cell nucleus, typical fatty degeneration of a whole group of cells is seen here and there. Pigment may be seen in those cells which contain parasites. The parasites appear to be in the capillary endothelium of the supporting tissue, and to a large extent in the stellate Küpffer cells. They are taken up, presumably, by phagocytosis. Changes in the interlobular connective tissue are not always to be found. Knot-like aggregations of white blood-corpuscles have been observed round the wall of the vessels (Nicolle). Venous or biliary cirrhosis is sometimes found (Gabbi, Abate).

Statham points out that one can see endothelial cells containing *Leishmania* between the columns formed by the hepatic cells. The endothelial cells are flattened out inside the blood-vessels. According to Christophers and others, the parasites are found in largest numbers in the endothelial cells inside the capillaries. According to Ledingham, the parasites are found in wandering leucocytes originating from the spleen.

According to Laveran and Havet the hepatic cells are the favourite position of the parasites in the liver, and they are found in much smaller numbers in the connective tissue, the plasma cells or the endothelial cells of the capillaries. They do not agree with Christophers as to the site of election of *Leishmania* in the liver in human kala-azar.

The author considers that the Leishman-Donovan bodies are mostly present in the Küpffer cells (Plate VII, fig. 3), agreeing with Christophers' observation. The accidental superimposition of parasites over liver cells or the apposition of parasite-laden Küpffer cells to the liver cells may give an impression of the parasites occupying the liver cells themselves.

Christophers points out that the capillaries of the lobules and the liver cells in certain zones are atrophied. The parasites are more often found in the periphery of the cells containing them than in their centre.

Shortt points out from his observations in experimental animals that the liver cells proper do not contain parasites. He also found an extreme degree of fatty infiltration and degeneration of the liver cells, and the liver tissue resembled the condition seen in phosphorus poisoning and in acute yellow atrophy. In infected hamsters there may be occasional parasitization of the liver cells (Meleney).

According to Rogers' statistics the liver was enlarged in 43 per cent. of his Assam cases, in 59 per cent. of the early European hospital cases, and in 75 per cent. of his cases at the Calcutta Medical College. It is usually very firm and hard in the more chronic cases. Rogers has described a form of intralobular cirrhosis in cases lasting two or more years :—

" It is characterized by an absolutely smooth surface of the organ, but on section the liver is found to be exceedingly tough, and so firm that digital pressure has no effect on it.

" Microscopically it shows very diffuse intracellular cirrhosis, in the

connective tissue of which shrunken parasites of kala-azar may still be visible with a high power.

"The liver cells are extremely atrophied, so that but little healthy secreting structure remains. In addition, multilobular cirrhosis is met with not very

FIG. 23. (1) Leishman-Donovan bodies inside liver cells; (2) Leishman-Donovan bodies in lymphatic glands; (3) Leishman-Donovan bodies in bone marrow. (From Statham's paper in the *Journal of the Royal Army Medical Corps.*)

rarely in old cases of kala-azar. At autopsy it is usually found to be associated with old dysenteric ulcers."

In some cases the author has found an extensive fatty degeneration of the liver with some pigmentation (fig. 23). Dilatation of the vessels is frequently seen and infarcts may be present.

Bone-marrow.—The marrow of the long bones becomes pink in colour, and in appearance resembles fœtal bone-marrow, and is somewhat diffluent. It is the elements forming the spongiosa which contain the greatest number of parasites (fig. 24). There are no marked changes to be observed either in the connective tissue or in the blood-vessels. The tunica media of the medium-sized vessels may undergo fatty degeneration (Visentini). According to Marchand and Ledingham, the cells of the small marrow veins contain no parasites. No deposit of pigment or hæmorrhage has been observed. Jemma and di Cristina found an increase in the number of lymphoid cells and a growth of the endothelium of the lymph channels and blood-vessels. "Clasmatocyte tissue" has been found in the bone-marrow of ribs in man.

FIG. 24. Section of bone-marrow showing the presence of Leishman-Donovan bodies inside endothelial cells.

(From a specimen kindly lent by Dr M. N. De of the Pathological Department of the Calcutta Medical College.)

Leishmania are found in the same cells of the bone-marrow as in the spleen. Statham found them in large and small myelocytes. They may also be found in the large mononuclear and polymorphonuclear leucocytes. According to Christophers they are mostly present inside the macrophages.

In experimental animals Shortt found them much less numerous in the bone-marrow than in either spleen or liver. The endothelial cells were greatly hypertrophied in affected areas and were crowded with parasites. Large parasite-laden macrophages were seen in intimate connection with the endothelium of capillaries and stroma cells.

The parasites retain their morphological characters longer in the bone-marrow than in other tissues after death.

Hu and Cash have found erosion of the inner table of the skull associated with hyperplasia of bone-marrow and extra-medullary deposit of blood on the surface of the dura mater in cases of kala-azar in China.

CHAPTER XIV.

TREATMENT.

THE TREATMENT of kala-azar, before the days of antimony, gave very indifferent results and need not be discussed here. Various drugs were given a trial, but no success was achieved. Reference may be made to the articles by Leishman and Gabbi in the earlier editions of Mense's TROPENKRANKHEITEN, and also to the older works of the present author on KALA-AZAR AND ITS TREATMENT, and to Laveran's LEISHMANIOSES.

The resemblance of the disease to malaria and African trypanosomiasis has led from time to time to the use of various treatments that have been adopted in these diseases. The various methods of treatment that were adopted before the days of antimony in the treatment of kala-azar may be summarized as follows: alkaloids, especially quinine in massive doses, leucocyte-increasing drugs, alkalies, bone-marrow and spleen extracts, vaccines, arsenic, mercury and other metals, X-ray, splenectomy, intravenous injection of antiseptics, &c.

Manson was the first to advise antimony preparations in the treatment of the disease. Vianna and Machado, in 1913, first used tartar emetic in the treatment of cutaneous leishmaniasis of South America, and obtained remarkably good results. di Cristina and Caronia used tartar emetic in the treatment of infantile kala-azar with satisfactory results. Their method of treatment was adopted by many workers in Italy, including, among others, Spagnolio, in Messina, Longo and Abate, in Catania, Jemma and his colleagues, in Naples. At the same time tartar emetic was beginning to be used in India in the treatment of kala-azar. As pioneers in this field, in the treatment of Indian kala-azar, the following may be mentioned: Rogers, Mackie, Hume, Castellani, and the author.

Rogers claims to have been the first to use it in the treatment of kala-azar. Castellani, however, has drawn the author's attention to the Report to the Advisory Committee for the Tropical Research Fund, 1914, by Castellani, in which his first note on the treatment of kala-azar with tartar emetic was published before Cristina, Caronia and Rogers published their results. He therefore claims to have been the first to use the drug in kala-azar. Also *vide* Laveran's LEISHMANIOSES.

Antimony can now be regarded as a specific in the treatment of the disease. The discovery of the best preparation has been the subject of extensive research, carried out by the present author during the last few years.

The subject of the chemotherapy of antimony cannot be discussed here in detail. The reader is referred to the papers written by the author on the

subject of "Chemotherapy of Antimonial Compounds," published in the *Indian Journal of Medical Research*. The following table gives the relative toxicity of some of the antimonial compounds investigated by the author :—

TABLE.

	Toxicity of the Sb content / 10 = 1 / (10 × Sb content % min. lethal dose)	Min. lethal dose in grms. for guinea-pigs per kg. of body weight.
Quinine antimonyl tartrate	$\frac{1}{30}$ ...	0·15
Sb_2O_3 dissolved in glycerine	$\frac{1}{25}$ ...	0·03
Ammonium antimonyl tartrate ...	$\frac{1}{23}$ ...	0·06
Strontium antimonyl tartrate ...	$\frac{1}{20}$ ...	0·055
Potassium antimonyl tartrate ...	$\frac{1}{20}$ ...	0·055
Calcium antimonyl tartrate	$\frac{1}{18}$ ...	0·055
Ethyl antimonyl tartrate	$\frac{1}{17}$ ...	0·045
Narcotine antimonyl tartrate ...	$\frac{1}{14}$ ...	0·085
Cinchonine antimonyl tartrate ...	$\frac{1}{30}$ ...	0·15
Sb_2O_3 dissolved in tartaric acid ...	$\frac{1}{25}$ ...	0·03
Urea antimonyl tartrate	$\frac{1}{21}$ ...	0·055
Ammonium antimonyl malate ...	$\frac{1}{20}$ ...	0·09
Sodium antimonyl tartrate	$\frac{1}{19}$ ...	0·055
Aniline antimonyl tartrate	$\frac{1}{17}$ ...	0·055
Lithium antimonyl tartrate	$\frac{1}{16}$ ...	0·04
Urea stibamine	$\frac{1}{259}$ ...	0·7
Stibamine	$\frac{1}{210}$ ...	0·5
Stib-hectine	$\frac{1}{170}$ ...	0·6
Stib-acetin	$\frac{1}{258}$ ...	0·7

The various antimonial preparations that have been or may be used in kala-azar may be conveniently divided under the following heads :—

(1) Aromatic antimonials: A new compound discovered by the author, and formed by the combination of p-amino-phenyl-stibinic acid with urea, urea-p-stibanilate, urea stibanilate, ammonium carbamino-stibanilate (named urea stibamine by the author); acetyl-p-amino-phenyl stibinate of sodium (stibacetin, stibenyl, Allen and Hanbury); sodium-p-amino-phenyl-stibinate or sodium stibanilate (named stibamine by the author); stibamine in combination with glucose (named glucose-stibamine by the author), or stibamine glucoside (Burroughs Wellcome and Co.); urea stibamine in combination with glucose (named glucose-urea-stibamine by the author); metachlor-p-acetyl-amino-phenyl stibinate of sodium (named chloro-stibacetin by the author), or stibosan (von Heyden); N-phenyl-glycine-amide-p-stibinate of sodium (a new compound described by the author and named stibiglycine-amide); a compound of unknown composition formed by the combination of p-arsanilic acid with an amine, and known as "693" von Heyden. Among the new aromatic antimonials prepared in the author's research laboratory and which are likely to be of therapeutic value are benzene-sulphonyl-p-amino-phenyl-stibinate of sodium (stib-hectine); urethane derivative of p-amino phenyl-stibinic acid; carbethoxy-p-amino-phenyl-stibinic acid; carboxy-methylene-oxyphenyl-4-stibinic acid; allyl-thio-carbamino-p-amino-phenyl-stibinic acid.

(2) The antimonyl tartrates: (*a*) Potassium antimonyl tartrate (tartar emetic), sodium antimonyl tartrate, sodium potassium antimonyl tartrate, hyper-acid antimonyl tartrate, ammonium antimonyl tartrate, lithium antimonyl tartrate, calcium antimonyl tartrate, strontium antimonyl tartrate, bismuth antimonyl tartrate, ethyl antimonyl tartrate, quinine antimonyl tartrate, cinchonine antimonyl tartrate, narcotine antimonyl tartrate. (*b*) The following new amino-antimonyl tartrates have been prepared in the author's research laboratory: Phenocoll antimonyl tartrate, anæsthesin antimonyl tartrate, novocaine antimonyl-tartrates, apothesine antimonyl tartrate, orthoform antimonyl tartrate, acriflavine antimonyl tartrate.

(3) Colloidal sulphide of antimony, colloidal metallic antimony, metallic antimony in a state of fine subdivision, antimony oxide dissolved in glycerine.

(4) Sodium antimony thioglycocollate and antimony thioglycocollamide.

(5) The antimonial malates.

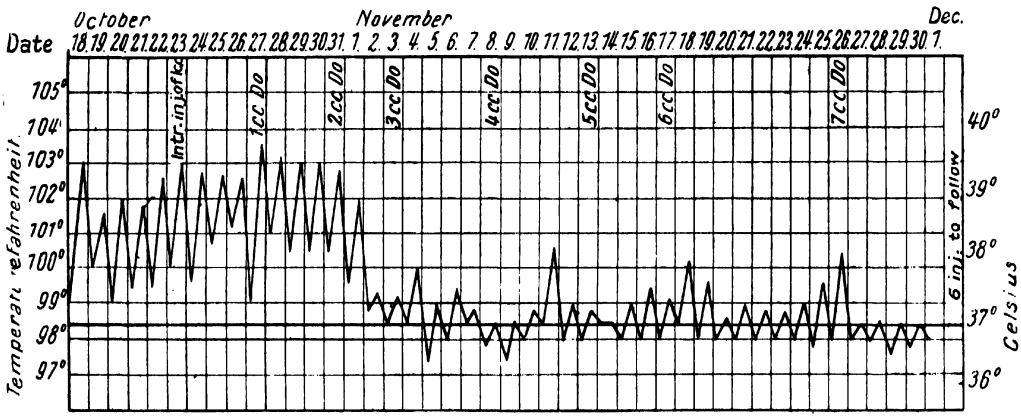


FIG. 25.—Temperature chart of a case of kala-azar cured by tartar emetic.

The antimonyl tartrates should not be considered as antimony salts of organic acids, as they are really salts of antimonyl tartaric acid. The pentavalent compounds of antimony should not be considered as equivalent to therapeutic aromatic antimonials, as they include inorganic salts of antimonic acid which have little or no therapeutic value.

The methods of administration of the antimonial preparations are: (1) INTRAVENOUS; (2) INTRAMUSCULAR; (3) INUNCTION; (4) ORAL; (5) RECTAL.

I. The INTRAVENOUS METHOD is the one most commonly employed, and gives the best results.

(*a*) The author in 1921 obtained very satisfactory results from the use of urea stibamine in the treatment of kala-azar. The compound was discovered by him in the course of research under the auspices of the Indian Research Fund Association. Full details regarding the use of urea stibamine in kala-azar and other aromatic antimonials are given later on.

(*b*) The two antimonyl tartrates that have been very commonly used are

tartar emetic and sodium antimonyl tartrate. The author has also used ammonium antimonyl tartrate. From the table given on page 111 it will be seen that this salt is the least toxic, and that the sodium and potassium salts are equally toxic. The dose of each of these salts for an adult is 2 to 5 c.c. of a 2 per cent. solution given intravenously twice a week. Some authors recommend the use of a 1 per cent. solution, but there is no advantage in this, nor is there any advantage in giving the injections at shorter intervals than twice a week. A higher concentration than 2 per cent. is not to be recommended.

The following precautions should be observed in carrying out a course of treatment with the antimonyl tartrates :—

If a severe reaction in the form of violent rigors, high fever and persistent vomiting or diarrhœa, should occur, the next dose should not be a larger one, nor should it be given after a short interval. In such cases the next dose should be smaller than the previous one, and given at an interval longer than that between the foregoing ones. If there is increase of œdema during the

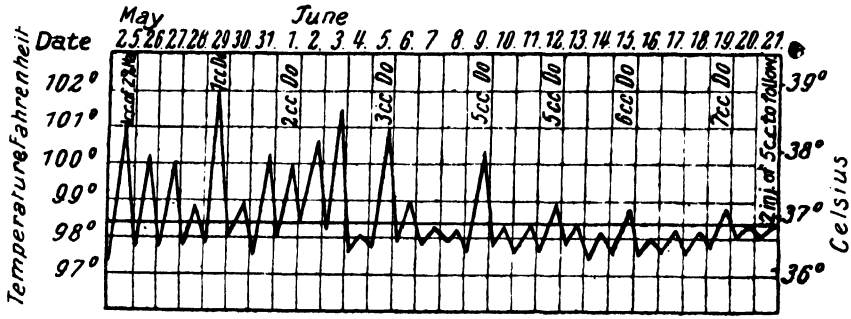


FIG. 26. Temperature chart of a case of kala-azar cured by sodium antimonyl tartrate.

course of treatment, then the dose should be diminished and given at longer intervals.

The treatment should be continued until the fever has been absent for a month. The blood-picture must be watched for a considerable time. The following factors should determine whether the treatment should be discontinued: A leucocyte count of 7,500 or more, without any tendency towards diminution, a steady gain in weight and improvement in the general condition, and in most cases reduction of the size of the spleen to normal. If possible, a spleen puncture and peripheral and spleen blood-culture should be made for absence of *Leishmania*.

Some observers hold that parasites may be present in the spleen even when the patient is clinically cured, and in some of these cases the parasites subsequently disappear spontaneously. It is risky, however, to stop the treatment if parasites are still present, as the possibility of a relapse remains.

The spleen is generally reduced to its normal size during the course of the treatment. A spleen that feels very hard, and clinically gives the impression

of having undergone fibrous change, may return to normal under antimony treatment. In other cases a slightly enlarged spleen may persist for a long time even after the cure has been established, and it returns to its normal size slowly. It may thus be concluded that while it is desirable that the spleen should be reduced to its normal size when the treatment is completed, it is not always necessary to continue the injections until this condition is reached. A fibrous spleen may remain enlarged after the patient has recovered. One must take all the above facts into consideration in determining whether the treatment should be discontinued or not.

Symptoms of intolerance have sometimes been met with after intravenous injections of tartar emetic or sodium antimonyl tartrate. Their solutions should not be sterilized in an autoclave, as toxic decomposition products may be thereby produced. It has been recommended to sterilize tartar emetic solution in flowing steam, on two or three consecutive days (Castellani and Chalmers), or by filtering it through a Chamberland filter. Neither of these precautions is necessary if one boils the solution for ten minutes.

It is desirable always to use a fresh solution. The author has shown that old solutions of tartar emetic or sodium antimonyl tartrate are more toxic to experimental animals than fresh ones, apart from any bacterial contamination.

Ammonium antimonyl tartrate has been used in the same way as tartar emetic, and the results seem to be satisfactory. Reference will be made to hyper-acid antimonyl tartrate under the heading of intramuscular injection.

(*c*) The author has used colloidal sulphide of antimony intravenously as well as subcutaneously. For an intravenous injection, 20 c.c. of a 1 in 500 solution may be given (Rogers). Rogers treated ten cases which were cured after injection of about 37 cgrm. of this preparation.

More recent investigations have shown that colloidal antimony sulphide does not produce uniformly good results.

(*d*) The author has used colloidal metallic antimony, and metallic antimony in a state of fine subdivision, intravenously with satisfactory results.

Preparation of colloidal metallic antimony has been described by the author in the *Lancet*, 1916, October 21.

Colloidal metallic antimony is brownish-red by transmitted light and black by reflected light, being in this respect similar to Svedberg's colloid.

It was at first thought that colloidal metallic antimony prepared by the author's method was a very stable substance, but subsequent observations showed that it became slowly transformed to Sb_2O_3.

Dose of colloidal metallic antimony : 0·001 grm. to 0·002 grm. given intravenously, or intramuscularly, on successive days.

(*e*) The author has used metallic antimony in a state of fine subdivision intravenously. Before the introduction of urea stibamine it was the most powerful antimonial preparation in the treatment of kala-azar, according to the author.

The dose of metallic antimony in a state of fine subdivision is $\frac{1}{4}$ to 1 gr. (0·016 to 0·065 grm.) intravenously.

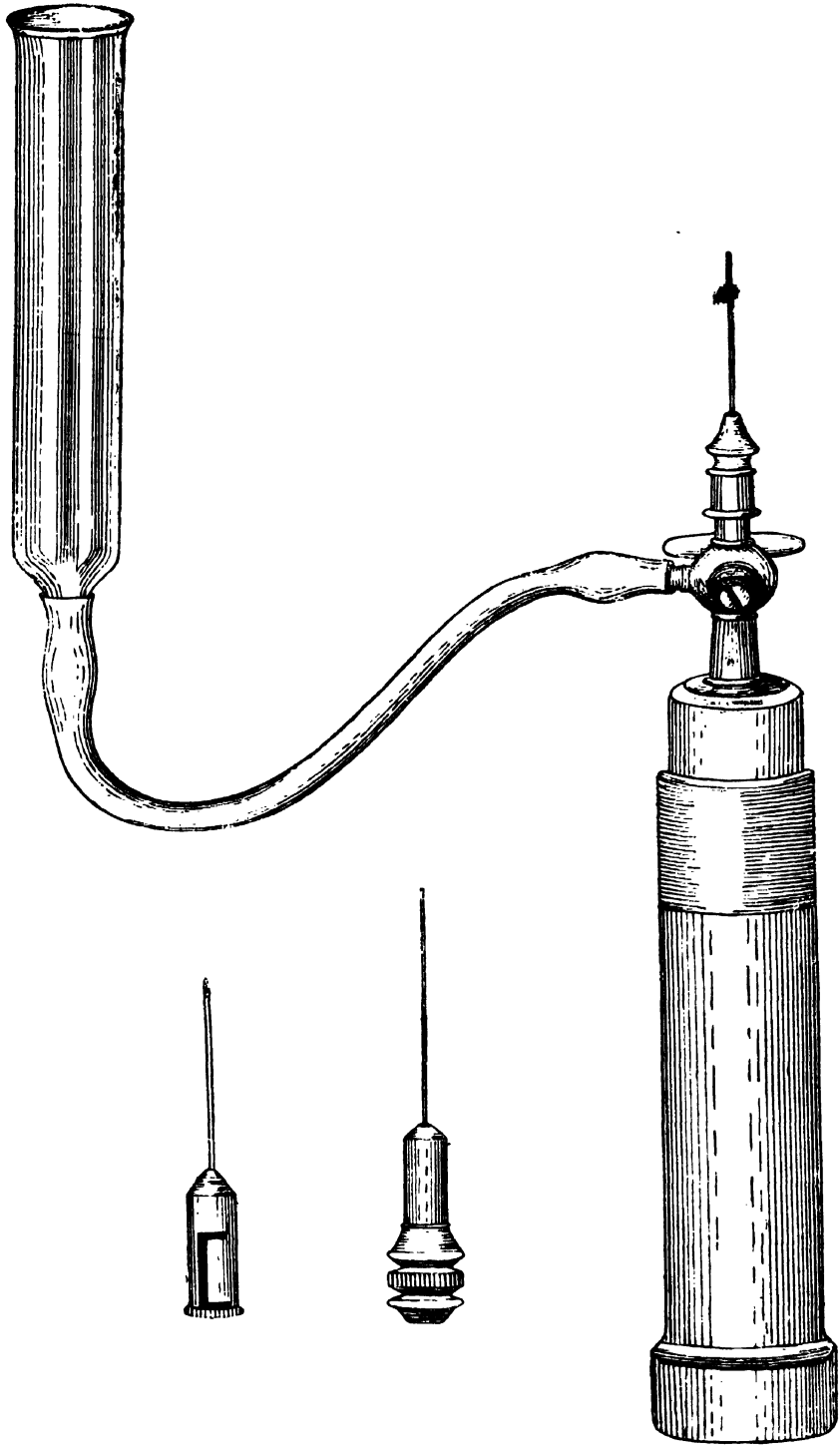


FIG. 27. Special apparatus used in the author's own method of intravenous injection of metallic antimony. (Original.)

(From the author's paper in the *Indian Medical Gazette*, December, 1915.)

A special apparatus has been devised by the author for intravenous injection of metallic antimony (fig. 27).

Technique of Intravenous Injection of Metallic Antimony.—The method of administration of metallic antimony is the author's own, and is described as follows (*Indian Medical Gazette*, December, 1915): An all-glass 10-c.c. syringe is fitted with a three-way stopcock, the remaining two ends of which are fitted to a platinum needle and a rubber tubing attached to the nozzle of a piece of glass tube respectively. A stout hypodermic needle, with a specially constructed blunt cannula inside, should be used preferably in place of an ordinary one. This cannula when pushed through

FIG. 28. A case of kala-azar before treatment with metallic antimony.

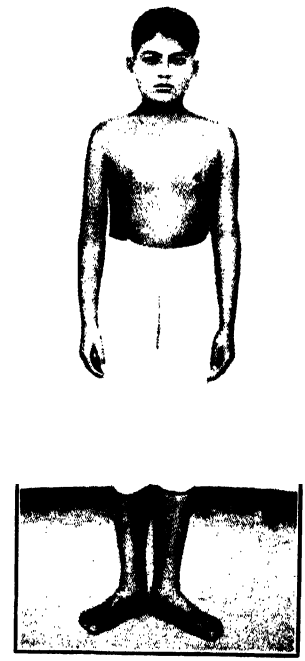


FIG. 29. A case of kala-azar after treatment with metallic antimony.

(Original.)

the needle will prevent the puncture of the vein a second time during the process of injection of the metallic antimony (see fig. 27). Formerly 1 gr. of metallic antimony was made into a thoroughly homogeneous paste with sterilized liquid glucose, in a glass mortar, and then mixed with 20 c.c. normal saline. (The glucose added is just sufficient to make a 5 per cent. solution with the 20 c.c. normal saline.) The stopcock is so arranged that the syringe may be made to communicate alternately with the needle or the glass tube by turning the stopcock. Half the suspension is now sucked into the glass syringe and, after being freed from any bubbles of air, it is injected into a vein of the fore-arm. The glass tube is now

filled with a portion of the remaining suspension, which is sucked into the glass syringe, and then the suspension is again injected. In this way the whole of the suspension is injected into the vein. Any sediment of antimony left inside the syringe is subsequently mixed with normal saline containing 5 per cent. glucose, and then injected into the vein. This process is repeated several times till no antimony is left inside the syringe. About 40 c.c. to 45 c.c. of normal saline are required to inject 1 to 1½ gr. of metallic antimony. The glucose has nowadays been completely dispensed with by the author, and the antimony is intimately mixed with normal saline by shaking it inside a stoppered glass phial. The antimony is sterilized by being dipped in absolute alcohol in a small stoppered phial, in which it is allowed to settle. After the antimony has settled down the alcohol is gently poured off. The antimony is subsequently transferred to the shaking phial, after being mixed up with normal saline. The patient should remain in the recumbent posture for half an hour after intravenous injection of metallic antimony.

Choice and Puncture of Veins for Intravenous Injection of Antimonial Solutions.—The patient must present himself for injection with an empty stomach. The presence of food in the stomach predisposes to vomiting. In the case of metallic antimony the patient is placed in the recumbent position, with his head on the same level with the feet. In other cases one may inject into a patient in the upright position, but the recumbent position is preferable, especially in debilitated patients. The choice of the vein need not be made until after compression of the arm; the venous circulation in the arm is stopped and the vessels dilated. (See fig. 30.) The cleaning of the skin is performed by friction of the field of operation with cotton-wool dipped in 90 per cent. alcohol. In some cases, after painting with tinct. iodine, the iodine stain is washed with absolute alcohol. This brings about perfect sterilization of the part. The median cephalic is usually the vein of choice. If the median basilic or cephalic vein cannot be made prominent, then one of the veins on the back of the hand or any other prominent vein should be tried. Concave veins, as are found over the inner side of the elbow of very muscular people, may be tried if the median basilic or cephalic vein does not become prominent. In cases in which the veins of the upper extremity are not prominent, the veins on the dorsum of the feet may be tried. There is one vein near the inner malleolus which may be particularly suitable for the purpose. In those cases in which the veins of the extremities are not available, the superficial veins of the abdominal wall may be tried. The veins are not always marked off on the skin by their blue colour. If they are big and thick-walled, their presence is perceptible by the prominences they form, even if they dip deeply into the subcutaneous tissues. Often, even by touching them with the point of the index finger passed over the surface of the skin, one perceives the existence of the blood-laden tube.

Puncture of the Vein.—The puncture of the vein does not present any difficulties if one knows the little details of the operation. If one wishes to succeed at once and without trouble, it is essential that the needle should catheterize the vein, i.e., that it should fit into it like a trochar into its outer

tube. To obtain such ideal catheterization it is of primary importance to fix the vein in such a manner that it does not run away before the needle. This is obtained by stretching the skin of the arm downwards with the point of the index finger or the thumb about 2 in. below the point aimed at. The stretching of the skin and the underlying tissue properly fixes the vein. This traction can also be obtained by forming an angle with the middle and index fingers of the left hand, this angle being bisected by the vein. Another means to immobilize the vein is to pass the left hand behind the arm of the patient and to stretch the skin on a vertical level with the thumb on one side. This position has the added advantage of fixing the arm as well, but it has the disadvantage of hiding the vein. After fixing the vein it has to be catheterized. One takes hold of the syringe with the needle and, pricking the skin over the vein, pushes the needle forward at a convenient angle. Instead

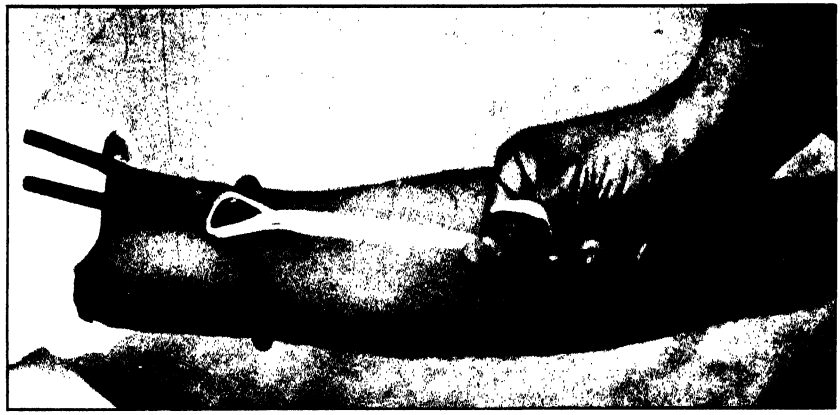


FIG. 36.—Author's method of compressing the veins at the bend of the elbow for the operation of vein puncture. A piece of rubber tubing is passed round the arm and then the two ends of the tubing are passed through one of the apertures forceps (Guy's pattern), in the way indicated in the figure. After p of the tubing and then clamping it, the pressure exerted in compress easily regulated.

of puncturing with the detached needle, the author considers it preferable to fix the needle to the syringe. The syringe is filled with the liquid to be injected, and after driving out the air until a drop of liquid appears at the point of the needle, the vein is punctured with the needle. At this moment the syringe is gently depressed and the piston is gently and slightly withdrawn. If any blood appears inside the syringe, then the needle is evidently inside the vein. To prevent the patient bending his elbow, it is always safe to hold the fore-arm a little above the wrist and exert gentle traction along its longitudinal diameter. At the termination of the operation the blood spilled on to the arm is washed with sterile water or swabbed with sterile cotton-wool. A pad of sterile cotton-wool is placed over the point of puncture and a little collodion is poured over it and gentle pressure applied.

II. INTRAMUSCULAR METHOD.

(*a*) The intramuscular injection of tartar emetic or sodium antimonyl tartrate is most painful, although not as painful as the subcutaneous injection. Castellani recommends the following formula :—

℞	Tartrate of antimony	8 gr. (0·52 grm.)
	Carbolic acid	10 min.
	Glycerine	3 drm. (11·4 grm.)
	Bicarbonate of sodium	¼ gr. (0·02 grm.)
	Distilled water	1 oz. (31 grm.)

The dose is ½ to 1 c.c. injected every other day intramuscularly into the gluteal region. Sodium antimonyl tartrate may be used in place of tartar emetic in the above formula and may perhaps be less painful.

(*b*) Antimony trioxide has been recommended for use in intramuscular injections in doses of 1 to 2 c.c. of a solution of 1 gr. (0·065 grm.) of the compound in equal parts of glycerine and water (50 c.c. each). The solution, which is probably a glycerate, is weak in its leishmanicidal properties.

(*c*) Colloidal sulphide of antimony in a 2 c.c. suspension in doses of 0·001 grm. has been used for intramuscular injections.

(*d*) Hyper-acid antimonyl tartrate (which is antimony trioxide dissolved in excess of tartaric acid) plus urethane (1 gr. of urethane added to 1 c.c. of a 1 per cent. solution of the Sb_2O_3) has been used by the author for intramuscular injections with very good results.

Dose : 0·05 to 0·1 c.c. of a 1 per cent. solution of the acid salt, calculated in terms of antimony trioxide dissolved in the tartaric acid. The injections are generally, but not always, painless.

(*e*) Urea stibamine has been given intramuscularly with beneficial results and frequently without much local irritation, the dose being 0·05 to 1 grm. dissolved in 1 to 2 c.c. of distilled water given on alternate days.

(*f*) Of the aromatic antimonials, intramuscular injections of acetyl-p-amino-phenyl-stibinate of sodium were first used by Caronia in the treatment of infantile kala-azar. He obtained good results from the administration of 3 to 10 cgrm. to children under 2 years, and 5 to 15 cgrm. to older children, on alternate days. Subsequent observations of Mackie and others, on the other hand, proved that the drug was toxic in its effects. The author has observed that in India it becomes toxic with age.

Some of the amino-antimonyl tartrates, prepared during the course of the author's research with the idea of using them intramuscularly, have been tried by him with variable results. They are less painful than the ordinary antimonyl tartrates.

III. INUNCTIONS.

In children and nervous patients an ointment of powdered metallic antimony in lanoline (5 or 10 per cent.) has been recommended to be rubbed in over the splenic region. In many cases it does not seem to make any impression upon the course of the disease. This treatment may, however,

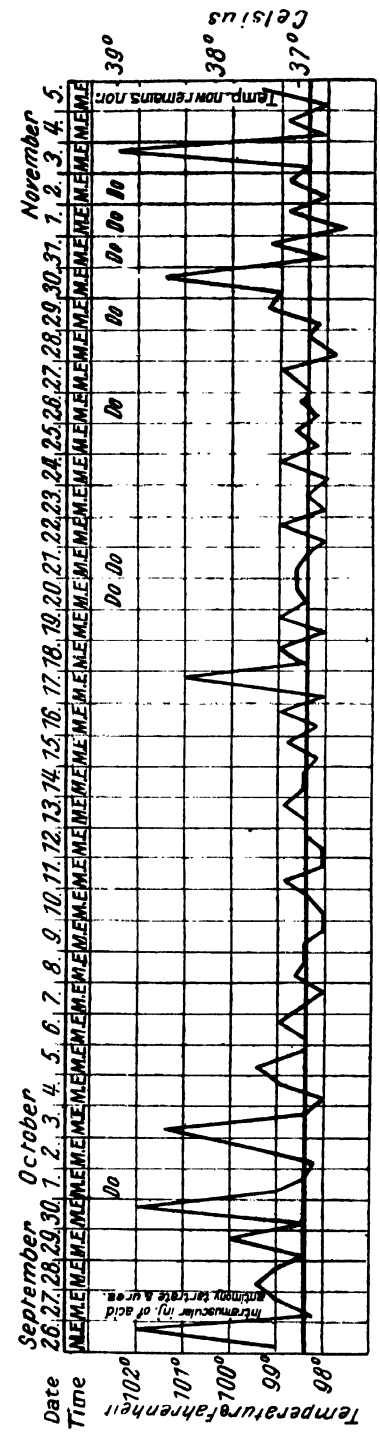


FIG. 31.—Temperature chart of a patient cured by intramuscular injections of hyperacid antimonyl tartrate with urethane.

supplement the intravenous injections. Ointment of metallic antimony in a state of fine subdivision is more easily absorbed and gives better results.

IV. ORAL METHOD.

(*a*) The following mixture may be given *per os* (Castellani) :—

R	Tartrate of antimony	5 gr. (0·325 grm.)
	Bicarbonate of sodium	30 gr. (2 grm.)
	Glycerine	1 oz. (31 grm.)
	Chloroform water	1 oz. (31 grm.)
	Water	ad 3 oz. (93 grm.)

The dose is one to two teaspoonfuls in water three times a day. According to the author, the therapeutic value of oral administration of the drug is weak.

(*b*) Bismuth antimonyl tartrate has been given orally by the author in a few cases. No conclusive results were obtained. Theoretically speaking, it should combine the therapeutic properties of bismuth and antimony.

V. RECTAL METHOD.—Sodium or potassium antimonyl tartrate has been administered per rectum, but this method has nothing to recommend it and is sometimes painful. Urea stibamine has also been administered by the same route.

In administering an antimonial preparation, such as tartrate of antimony or sodium antimonyl tartrate, it must be remembered that the concentration of antimony in the blood has to be high enough to destroy the *Leishmania*. It appears that, generally speaking, such a concentration can only be obtained by the administration of the above compounds by the method of intravenous injection, as sufficiently large doses cannot be administered by any other route without giving rise to distressing local symptoms.

The following symptoms of intolerance may follow intravenous injection of tartar emetic or sodium antimonyl tartrate :—

Rigors and rises of temperature, which may sometimes be severe. Occasionally the patient complains of intense headache lasting for some days, pain in the gums, vomiting, troublesome cough, diarrhœa or a dysenteric condition, which may sometimes be severe and alarming. Sometimes the patient complains of a metallic taste in the mouth, and there is a disinclination for food, and vague abdominal discomfort. Rarely the patient complains of intense abdominal pain, especially over the splenic and hepatic regions, lasting for some days, or in the region of the kidneys, associated with dysuria. In some cases a bronchitic condition sets in which lasts for some days.

In some cases œdema of the extremities, or an increase of œdema, if already in existence, makes its appearance after the first few injections. In a few cases a peculiar suffocating feeling or a feeling of spasm of the glottis occurs after the injection. Sometimes symptoms of cardiac depression, with a feeble pulse, may appear after the injection, but this is rare.

In one case, high fever with violent fits set in after injection of 10 c.c. of a 2 per cent. solution of antimonyl sodium tartrate. The patient remained comatose for two days, but afterwards recovered.

Jaundice may appear in some cases after intravenous injection of antimonial preparations. Herpes may appear occasionally.

Sometimes the patient complains of pain in the joints which may be severe and last many days. These joint pains generally come after the treatment has been continued for some time.

In a few cases the patients developed pneumonia shortly after injections of sodium antimonyl tartrate or tartar emetic; the same condition was observed in one case after intravenous injections of metallic antimony. It seems probable that injections of antimonial preparations predispose to attacks of pneumonia. Greatest care should be taken not to expose cases of kala-azar to colds or chills during antimonial treatment.

If the patient is suffering from bronchitis, it is advisable to postpone antimonial treatment, or if bronchitis develops during the course of treatment of the disease, antimonial medication should be suspended for some time till the complication disappears.

In very rare cases, symptoms of paraplegia may set in during treatment with sodium antimonyl tartrate or tartar emetic.

On very rare occasions death from extreme dyspnœa has taken place suddenly and unexpectedly after injection of these salts, though several injections may have been given previously without any untoward symptoms.

The therapeutic value of the antimonyl tartrates compared :—

Thomson and Cushny observed that sodium and potassium antimonyl tartrates were equally efficacious in the treatment of sleeping sickness. The author considers that they are also of the same value in the treatment of kala-azar. Others hold that the sodium salt is the more efficacious of the two. They give rise to much pain and inflammation when injected subcutaneously or intramuscularly, and sloughs may form, especially after subcutaneous injections. According to Plimmer and Fry, lithium-antimonyl tartrate is the most efficient trypanocide among the inorganic antimonyl tartrates. In experimental animals it gives much better results when given subcutaneously than the corresponding sodium or potassium salt. Among the organic antimonyl tartrates, Thomson and Cushny considered that ethyl antimonyl tartrate was the most efficient trypanocide of all the tartrates, and though it was more poisonous than the potassium or sodium salts, the difference between the effective trypanocidal dose and the lethal dose was not less than that of the other antimonyl tartrates, and the optimum dose corresponded to Browning's therapeutic dose, being two-thirds of the maximum dose that average animals tolerate. In kala-azar the author has found the drug to be so toxic that he gave up its use. A new method of preparing the compound has been described by the author. Quinine antimonyl tartrate has been found by Fargher and Gray to be the least toxic of all the antimonyl tartrates,and this has been confirmed by the observation of the author. The author has no experience of the drug in the treatment of kala-azar. As stated before, ammonium antimonyl tartrate is less toxic than the sodium or potassium salt. It is equally efficacious in the treatment of that disease.

In India sodium antimonyl tartrate is the most popular preparation of all the antimonyl tartrates that have been used in the treatment of kala-azar.

Bismuth antimonyl tartrate and urea antimonyl tartrate have been prepared for the first time in the author's research laboratory. The former contains both bismuth and antimony, and should combine on theoretical grounds the therapeutic value of both the elements.

Urea antimonyl tartrate has been used with success in a few cases.

It may be stated here that sodium antimony thioglycocollate and antimony thioglycocollamide have given good results in the treatment of experimental trypanosomiasis. These may be tried in the treatment of kala-azar.

AROMATIC ANTIMONIALS IN THE TREATMENT OF KALA-AZAR—UREA STIBAMINE AND OTHER ORGANIC PENTAVALENT ANTIMONIALS.

It will not perhaps be out of place to give here a short history of the introduction of aromatic antimonials in the treatment of Indian kala-azar.

The chemotherapy of aromatic antimonials in kala-azar infection has been the subject of research by the author for many years. In 1920, shortly after he had been financed by the Indian Research Fund Association for carrying on researches into the treatment of kala-azar, *acetyl-p-amino-phenyl stibinate of sodium* and *amino-phenyl-stibinate of sodium* were prepared for the first time in India in his laboratory in the Calcutta Campbell Medical School, and he immediately brought to the notice of the Government, the Governing Body of the Indian Research Fund Association, and the then Secretary of the Calcutta School of Tropical Medicine, the possibility of the potentialities of these compounds in the treatment of Indian kala-azar, his conclusions being based on theoretical grounds, from an analogy of the value of the corresponding compounds of arsenic, namely, *ars-acetin* and *atoxyl*, in the treatment of certain protozoal diseases.

The acetyl compound (= stibacetin, stibenyl) was used more or less successfully outside India in the treatment of kala-azar and other forms of leishmaniasis (Caronia, Kharina-Marinuchi, Spagnolio). Manson-Bahr successfully used it in a case of kala-azar. Early in 1921 the author discovered that urea could combine with stibanilic acid and that the resulting compound surpassed all his expectations in its value in the treatment of kala-azar. Its introduction by the author and his researches into the chemotherapy of antimonial compounds in kala-azar infection opened up a new vista in the treatment of the disease in India, by means of therapeutic aromatic antimonials.

Urea stibamine (*ammonium carbamino-stibanilate, urea-p-stibanilate or urea-stibanilate*).

TREATMENT OF KALA-AZAR WITH UREA STIBAMINE.—REVIEW OF THE MOST IMPORTANT PAPERS ON THE USE OF THE COMPOUND. METHOD OF ADMINISTRATION—ITS DOSAGE—INDICATIONS AND CONTRA-INDICATIONS—ADVANTAGES.

Until recently tartar emetic or sodium antimonyl tartrate was extensively used for the treatment of kala-azar in India. These are now being completely

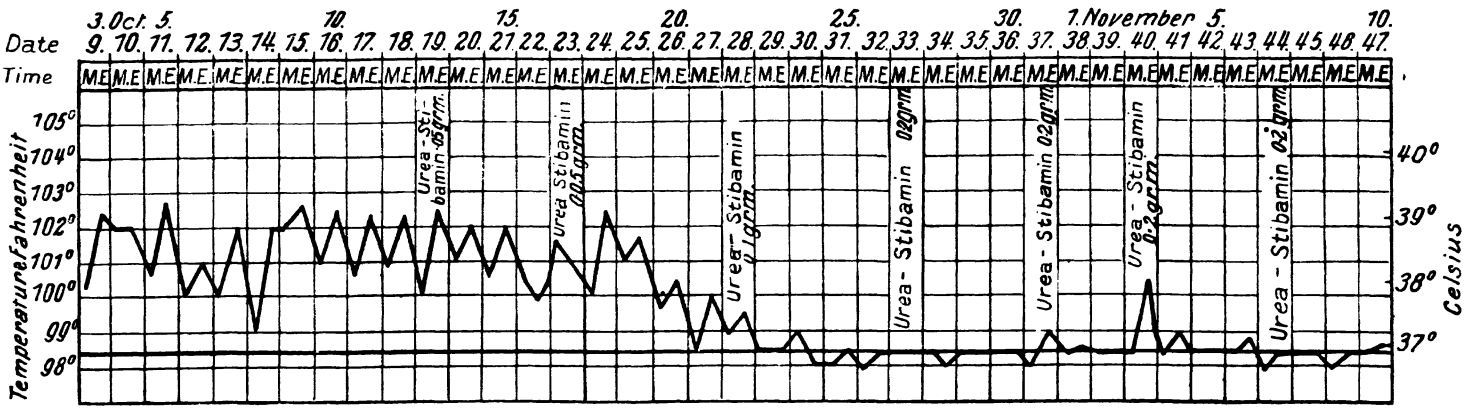


FIG. 32.—Temperature chart of a case cured by intravenous injections of urea stibamine.

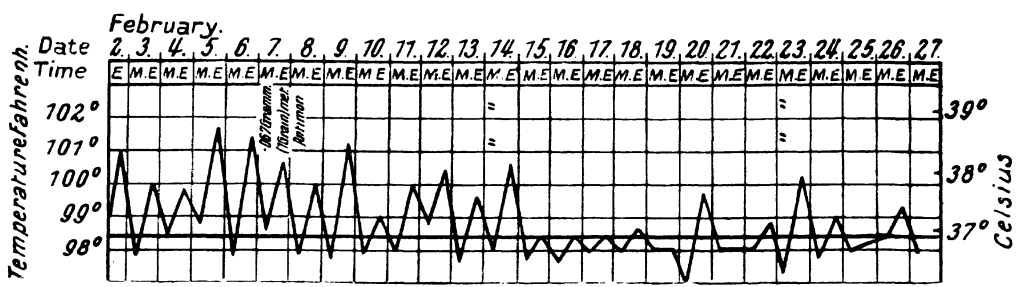


FIG. 33.—Temperature chart of a case cured by intravenous injections of metallic antimony. (Original.)

replaced by urea stibamine, on account of its high therapeutic value and marked superiority over the antimonyl tartrates.

The compound formed by the combination of urea with stibanilic acid was named by the author urea stibamine. The medical profession in and outside Calcutta soon came to recognize its value from reports of cases treated with the drug in the wards of the author in the Campbell Hospital, long before the results of his observations were published. His first series of successful cases were published in October, 1922. In 1923, Shortt and Sen in Assam reported having obtained more brilliant results with this compound than those obtained by the author in his first series of cases.

The Governing Body of the Indian Research Fund Association quickly recognized its value from reports of the author working in Calcutta, and also of those obtained from Shortt and other successive Directors of the Pasteur Institute, Shillong, from Christophers, Director of Kala-azar Commission, who reported from his experience in Assam about its remarkable efficacy, from medical officers of tea estates in Assam, and from the Government of Assam. In Calcutta its value was recognized by the physicians of the Calcutta Medical College Hospitals and successive superintendents of the Calcutta European Presidency General Hospital. In these institutions it was quickly introduced and extensively used with most brilliant results. Its reputation quickly spread all over Assam, Bengal, Bihar and Orissa, and to more distant places in India such as Madras, Sanawar, Simla Hills, and other places too numerous to mention, and every observer who used the drug was convinced of the great advance made by its discovery in the treatment of kala-azar.

Remarking on its therapeutic value, the Secretary, Scientific Advisory Board, Indian Research Fund Association, wrote as follows: "Both the Director of the Kala-azar Commission and Lieut.-Col. E. D. W. Greig consider that this drug has a highly specific action in kala-azar, and its value has been abundantly testified to by those who have tried it, both in so far as it shortens the period of treatment and in so far as it seems able to cure intractable cases or cases which are resisting the antimony treatment."

The author, while discussing with the Director of the Calcutta School of Tropical Medicine about the therapeutic value of urea stibamine, soon after its discovery, pointed out and suggested to him the possibility of obtaining therapeutic aromatic antimonials from Chemisch Fabrik von Heyden, the only compound of that nature then available in England being stibenyl, and subsequently "471" von Heyden was introduced in this institution for the treatment of kala-azar.

Of all aromatic antimonials that have been used up to the present time, the most extensive literature has been published on urea stibamine. The reports consist of cases treated by different observers, and under different conditions, and are therefore most valuable, there being no personal element in the choice of cases.

A Short Review of the Most Important Papers on the Use of Urea Stibamine.— Urea stibamine, discovered by the writer in the course of research under auspices of the Indian Research Fund Association, has been given extensive

trials in the treatment of kala-azar by different observers during recent years. A review of the most important published papers on the compound and of a few unpublished ones is given below.

Chemical Constitution and Toxicity.—The constitutional formula of urea stibamine is $NH_2CO.NH.C_6H_4SbO.OH.ONH_4$ (*Indian Journal of Medical Research*, October, 1924). The $NH_2CO.$ group shows that it is allied to tryparsamide, an arsenic compound, upon which the future conquest of African sleeping sickness depends, as pointed out by Dale. Originally, it was considered to be urea salt of stibanilic acid.

LETHAL EFFECTS PRODUCED FROM THE ADMINISTRATION OF A 2-PER-CENT SOLUTION OF UREA STIBAMINE TO GUINEA-PIGS BY INTRAMUSCULAR INJECTION. (Brahmachari.)

Dose per kg.	No. of guinea-pigs used	No. died	Remarks
0·7 grm.	4	4	M.L.D.
0·65 ,,	3	2	Maj.L.D.
0·6 ,,	4	2	...
0·5 ,,	2	1	...
0·45 ,,	4	1	...
0·4 ,,	4	1	...
0·35 ,,	4	*Nil.*	M.T.D.

The maximum tolerated dose of urea stibamine per kilogramme of body weight is twenty-three times that of tartar emetic in the case of the guinea-pig. The effective dose of urea stibamine in the treatment of kala-azar is five-sevenths the tolerated dose for the guinea-pig, while in the case of tartar emetic, it is eight times the tolerated dose for the same animal. Urea stibamine is therefore a much safer antimonial for use in the treatment of kala-azar than tartar emetic or other antimonyl tartrates.

Value of Urea Stibamine in the Treatment of Kala-azar.—I. The first paper on the subject was published in October, 1922 (*Indian Journal of Medical Research*), in which Brahmachari published a series of eight cases of kala-azar successfully treated with urea stibamine. In this paper the toxicity of the drug was fully tested, which is summarized above.

II. The second paper on the subject was that of Major Shortt, I.M.S., Special Kala-azar Officer, Pasteur Institute, Shillong, and Dr. R. T. Sen, published in the *Indian Medical Gazette*, July, 1923. In this paper they described a series of five cases of kala-azar treated with urea stibamine with most encouraging results. They stated that its advantages were: (*a*) The short course, occupying only two to three weeks necessary for a complete cure; (*b*) the rapidity with which the symptoms of the disease disappear; (*c*) the fact that no symptoms of intolerance were met with in any of their cases.

The cases were particularly severe, as the following remarks of the authors will show :—

1st case.—Case on admission was very ill with marked œdema of legs and great weakness. 2nd case.—Case on admission, weak with œdema of feet. 3rd case.—Case on admission, emaciated, weak and very anæmic. 4th case.—Case on admission, extremely weak with very severe bronchitis. Parasites very numerous.

III. The next paper was a second contribution by Brahmachari, published in the *Indian Journal of Medical Research*, October, 1923, in which he

recorded his further observations on the treatment of kala-azar with urea stibamine. A series of nine fresh cases, successfully treated, were noted with observations on others previously recorded. The latter had shown no relapse after one year after treatment was stopped, the patients being kept under observation for the period.

IV. The fourth paper was that of Shortt and Sen, published also in the above issue of the *Indian Journal of Medical Research*, October, 1923. It consists of records of twenty-one *consecutive* cases of kala-azar treated with urea stibamine, one of which was a very severe case of fulminating type with intense infection. All the cases were cured.

The following observations were made by Shortt and Sen in connection with their paper : (*a*) *Tolerance to the drug.* Even in those patients to whom the largest total quantity of urea stibamine was exhibited no symptoms of intolerance appeared. The greatest quantity administered to any case was over 5 grm. The result was in marked contrast to that obtained with the usual antimony salts, where, in a large percentage of cases, symptoms of intolerance, in the form of joint pains, &c., appear after greater or lesser amounts of the particular salt in use. (*b*) *Amount of urea stibamine and time required for sterilization.* This varied with the severity and previous duration of the case. Uncomplicated cases of average severity, especially if of recent origin, were usually cured by 2 grm. of the preparation. Administered on alternate days, this represented a period of about twenty days. A few cases were cured more rapidly. Long standing cases and some with very acute infections of the fulminating type took a longer period and larger total quantities before sterilization was established. Several of the cases were those which had resisted completely treatment by sodium and potassium antimony tartrate, yet in no single case did the authors fail to procure sterilization by the use of urea stibamine. (*c*) *Absence of local irritation.* Urea stibamine injected subcutaneously produced little, if any, local irritation. The importance of this, especially in the treatment of young children, will be realized by anyone who had seen the results of the escape of antimony salts into the tissues, when intravenous administration has been attempted unsuccessfully by an unskilled operator. This absence of local irritation raises the question of the possible use of this preparation by subcutaneous or intramuscular administration in special cases. This method is now being given a trial by us, with apparently successful results. (*d*) *Rapidity of action.* The amelioration of physical signs and symptoms was more rapid than with the ordinary antimony salts. Reduction of the temperature, diminution in the size of the spleen, and generally improved condition of the blood, were more rapidly achieved. (*e*) *Action in producing leucocytosis.* In many of the cases urea stibamine had a most beneficial action in producing a rapid improvement in the total leucocyte count. This effect, always a favourable sign in kala-azar, was in many cases most marked, and would alone be a point in its favour. (*f*) *Importance of treating cases as early as possible.* The reasons are twofold : (1) Early cases were found to yield more readily and rapidly to treatment, and the affected tissues were able completely to recover their tone and normal

functions. (2) Very old standing or advanced cases might have their tissues and organs permanently damaged by the disease to such an extent that, even although sterilization as regards parasites was effected, yet their functioning would never be recovered and the duration of life would necessarily be greatly curtailed.

The average amount of urea stibamine used per case was found to be 2·6 grm. The average number of injections required for, and the average period occupied in, actual treatment with the drug were found to be twelve injections and thirty-two days respectively. When this is compared with the thirty injections and ninety days usually required for a minimum full course of sodium antimony tartrate, the advantages of the use of urea stibamine will be obvious, and the more so, as it is apparently capable of sterilizing some of the cases which definitely resist treatment by the usual salts of antimony. This latter fact was evidenced in one case which had received 653 c.c. of 1 per cent. solution of sodium antimony tartrate with no apparent benefit, but was cured with urea stibamine after only seventeen injections.

The authors concluded that urea stibamine was by far superior to any other antimony preparations in general use.

V. The fifth paper was that of Brahmachari, published in the *Indian Journal of Medical Research*, April, 1924, which gives a series of nine consecutive cases of kala-azar *resistant* to antimony tartrates cured with urea stibamine.

By *refractory or resistant* cases of kala-azar were meant cases which resisted treatment with 2 grm. or more of sodium or potassium antimonyl tartrate given intravenously in the routine form of treatment of the disease extending over a period of two months and a half to three months or more.

The conclusions were: (1) The curative value of urea stibamine and its superiority over the antimonyl tartrates were established; (2) refractory cases resistant to antimonyl tartrates yielded to urea stibamine; (3) the short course of treatment and the lesser number of injections required in bringing about a complete cure were striking; (4) no relapse was met with among the cases that had undergone complete treatment with urea stibamine; some of these were under the author's observation for nearly two years after completion of treatment; (5) no relapse was met with among the resistant cases that were subsequently cured by urea stibamine. Some of these cases were under the author's observation for about a year; (6) the possibility of using urea stibamine intramuscularly was suggested.

VI. The next paper on urea stibamine is that of Dr. Percy Foster, Badlipar Tea Estate, showing the value of the drug in the treatment of kala-azar under tea garden conditions in a series of twenty cases (*Indian Medical Gazette*, August, 1924).

Remarks of Dr. Percy Foster on the progress of his cases during treatment with urea stibamine: The rapidity of the diminution of the size of the spleen was most marked in a very large percentage of his cases. The manner in which the fever was checked after the second injection was also a point well illustrated in a large number of the cases. Dr. Foster further remarked that all

his cases were subsequently inspected and showed a marked improvement in general health and weight. All the cases were cured, except one which died. Three of the cases treated were cases of relapse.

VII. Dr. Foster's work on the use of urea stibamine was continued by his successor, Dr. K. L. Banerjee.

Remarks of Dr. Banerjee : "Urea stibamine is a remarkable drug and a certain cure for kala-azar. The great advantage of this new drug is the *short course of treatment*. Generally it takes three weeks to cure a patient with this drug if the injections are given on alternate days, and I have practically seen cases being cured within eight days after four injections only. What a powerful drug it is as compared with other antimony preparations which take three to four months to cure a patient! The cases which resist treatment with ordinary antimony preparations are cured with urea stibamine. I have seen cases, which got about 400 c.c. of 1 per cent. solution of sod. ant. tart. with no effect, cured by urea stibamine within three weeks. Fever subsides after one or two injections and the general health improves rapidly, more so after the injections are over. My treated cases are now enjoying excellent health, and those who had some enlarged spleen at the end of the treatment have their spleen reduced to normal size. I have not any bad effects after urea stibamine injections. No joint pains are complained of by the patients. Children tolerate the drug very well, and I have given them in increasing doses from 0·05 grm. to 0·25 grm. without any bad effects. If a little of the solution goes by chance into the tissues there is no marked pain or bad effect, but only a slight inflammation is noticed which subsides within two or three days. I have given urea stibamine intramuscularly in the gluteal region to a child without any evil effects, and the child was subsequently cured."

The cases treated by Foster and Banerjee belonged to Hantley Tea Estate, Assam. The total number was sixty-seven. Leaving one case that was admitted into hospital in a moribund state, and another, aged 35, weighing only 73½ lb. and having lung complications before admission, the percentage of cures was 98 per cent., the proof of cure being negative results from spleen puncture and disappearance of the symptoms at the time of discharge from hospital and subsequent following up of the cases after discharge from hospital. Of the cases treated, three required four injections, twenty-two between four and eight for complete cure, and the average for all the cases treated was ten.

VIII. The next paper is that of Dr. Dodds Price, Medical Officer, Solana Tea Estate, on cases of kala-azar showing little or no improvement with sodium antimonyl tartrate, subsequently cured by urea stibamine (*Indian Medical Gazette*, September, 1924).

The present writer was very kindly furnished by Dr. Dodds Price with notes of thirty-seven such cases, the first eight having been published. Besides, Dr. Dodds Price sent him notes of fifteen other cases with the following remarks : "All these were taken in the *early* stages of the disease, and the results have been uniformly successful, and no case has required more than ten injections, the average working out at seven."

IX. The next paper is that of Brahmachari on the value of urea stibamine in the treatment of early kala-azar, published in the *Indian Journal of Medical Research*, October, 1924.

TABLE SHOWING THE VALUE OF UREA STIBAMINE IN THE TREATMENT OF EARLY KALA-AZAR.

	Cal. Med. Coll. Hosp. Barnardo, McCay, Author						Pasteur Inst., Shillong. Shortt, Greig and Kundu					Hantley Tea Estates. Foster and Banerjee		Author and Maity		
Duration of illness in days	18	4	120	8	10	21	135	90	120	60	90	7	15	90	150	150
Amount grm. after which cult. neg.	0·35	0·3	0·4	0·4	0·3	0·2			0·7	0·7	0·95			0·5	0·2	0·25
Total amount grm. given	0·35	0·3	0·4	0·5	0·5	0·2	0·7	0·75	0·7	0·7	0·95	0·35	0·5	0·5	0·2	0·35
Period of treatment in days	14	10	12	10	14	5	7	7	5	5	7		7	4		

Brahmachari concluded as follows: "Urea stibamine cuts short the course of kala-azar to a remarkable degree, if administered in its early stages. The same conclusions have been arrived at from observations made by different observers in different places. Its beneficial effects in its early stages are noteworthy.

"It is evident from an economic point of view and in the interests of the miserable sufferers, that the use of urea stibamine in the early stages of the disease cannot be over-emphasized."

X. The next paper is that of Dr. Michael, Medical Officer, Agricultural Research Institute, Pusa, published in the last edition of the author's KALA-AZAR AND ITS TREATMENT (1925).

Dr. Michael concluded as follows: (1) The most important action of urea stibamine appears to be the rapidity with which it checks the toxæmia of kala-azar, as evidenced by rapid reduction of fever. This opens out great possibilities in the treatment of the disease with urea stibamine in its early stages, the advantages to be gained in an epidemic, for instance, being immense. (2) Urea stibamine is practically non-toxic in its action and does not set up irritation of the vital organs. (3) It causes a much more rapid reduction of the spleen than tartar emetic or sodium antimony tartrate. (4) The advantages to doctor and patient alike of a very short course of injections for cure need no comment.

XI. The next paper on urea stibamine is a paper of Brahmachari and Maity, containing a very interesting series of kala-azar cases cured with urea stibamine in thirty-two hours to seven days (published in the *Indian Journal of Medical Research*, April, 1925). Some of these cases had an *intensive* course of treatment consisting of multiple injections of urea stibamine during twenty-four hours.

TABLE SHOWING A FEW CASES CURED BY INTENSIVE TREATMENT WITH UREA STIBAMINE.

Duration of illness in months	3	5	3	7	2	3	1	5	—		
Amount in grammes after which blood-culture negative	0·15	0·85	0·3	0·65	0·65	0·8	0·4	0·4	0·75	1·4	0·65
Total amount grammes injected	0·35	0·85	0·3	0·75	0·85	0·9	0·4	0·4	1·35	2	1·15
Period of treatment in hours	120	72	72	30	58	90	54	32	168	288	192
Days of observation after treatment	30	25	15	25	6	12	18	33	40	55	210

XII. The next paper is that of Captain Kapur, I.M.S., Resident Physician, Medical College Hospital, Calcutta, on "Observations on the Treatment of Kala-azar with Urea Stibamine in the Medical Out-Patient Department of the Medical College Hospital, Calcutta" (*Indian Medical Gazette*, vol. lx, 1925). They include a series of cases which were found to be definitely resistant to treatment with antimonyl tartrates.

Observations.—(1) The course of treatment with urea stibamine for bringing about cure in kala-azar is remarkably short. In some of the cases cure took place in only seven days. In case of antimonyl tartrates, the course of treatment frequently extends to more than three months.

(2) The average number of injections required for a course of treatment with urea stibamine is seven, while in the case of antimonyl tartrates it frequently amounts to thirty or more.

(3) The beneficial effects obtained with urea stibamine in cases resistant to the antimonyl tartrates are striking.

(4) Jaundice and albuminuria are no contra-indications to the treatment of kala-azar with urea stibamine. Such cases are best treated with small doses slowly increased.

(5) No signs of intolerance were observed in any of the cases. Sometimes a reactionary fever took place after injection which subsided after a few hours.

(6) Cases in which signs of intolerance showed themselves after injection of sodium antimonyl tartrate bore urea stibamine very well.

(7) The most striking results with urea stibamine are (*a*) rapid disappearance of the spleen; (*b*) rapid subsidence of the fever; (*c*) early disappearance of leucopenia; and (*d*) short course of treatment required for a complete cure.

XIII. The next paper is that of Major Shortt and Sen, being entitled, "Final Report on the Use of Urea Stibamine in Kala-Azar," published in the *Indian Journal of Medical Research*, October, 1924.

The following conclusions are quoted from that paper: "We consider that the value of urea stibamine has been established as the most efficient drug at present in use for the treatment of Indian kala-azar. The conclusion is based not only on a series of cases of which we have published the details, but in addition on experience gained in many other cases, both Indian and European, which have passed through our hands or which have been treated

with urea stibamine under our direction, a number totalling nearly one hundred cases."

XIV. The next paper is that of Colonel Greig, Director, Pasteur Institute, Shillong and S. Kundu, published in the *Indian Journal of Medical Research*, April, 1925. It gives the records of fifty-one cases cured by urea stibamine with complete sterilization of the internal organs.

XV. The next paper is that of Kundu, published in the December issue of the *Indian Medical Gazette*, 1925. It shows the value of intramuscular injection of urea stibamine in the treatment of kala-azar.

XVI. The stability of urea stibamine. Major Shortt, Director of Kala-azar Commission, has observed that urea stibamine stored in sealed ampoules for six months without any other precautions showed no deterioration or other change either in physical character or therapeutic properties.

A paper on the toxicity and therapeutic value of old samples of urea stibamine, by Brahmachari and Maity, has been published in the *Indian Journal of Medicine* (September) and *Calcutta Medical Journal* (August), 1926. They conclude as follows: "Urea stibamine has been found to remain perfectly stable up to the time of our observation, i.e., January, 1926, after having been preserved in sealed ampoules since 1922 under ordinary conditions in Calcutta and elsewhere without any deterioration, i.e., four years after the samples had been preserved. The therapeutic properties of these old samples remain fully intact and these have been used in the treatment of kala-azar without any untoward results. There is no difference in toxicity between old and new samples of urea stibamine."

Method of Administration—Dosage—Indications and Contra-indications—Advantages.

Preparation of Solution and Mode of Administration.—It should be used intravenously dissolved in cold sterile water. The solution should not be boiled. For making the solution dissolve 0·05 grm. and 1 grm. in 1 c.c. of water, 0·15 grm. and 0·2 grm. in 2 c.c. of water. Always use a fresh solution.

Dosage.—It is always desirable to start the treatment as soon as diagnosis of kala-azar is made, as the cases treated in the early stages of the disease frequently show the speediest recovery.

For adults, begin with 0·05 grm. and increase by 0·05 till the dose reaches 0·2 grm., which is generally the highest dose. Any one of the doses may be repeated, according to the discretion of the physician, before proceeding to the next higher dose. (Some observers begin the treatment of the disease with 0·15 grm. or 0·2 grm. with satisfactory results.) Generally, the injections are given twice a week, though more frequent injections also may be given. The injections should be continued till the symptoms of the disease disappear.

For children about 5 years of age, begin with half the initial dose for adults, i.e., 0·025 grm. and increase up to 0·05 grm. or 0·1 grm. Any one of the doses may be repeated, according to the discretion of the physician, before proceeding to the next higher dose.

For children about 1 year of age, begin with one-fifth the initial dose for adults and increase up to 0·025 grm. or 0·05 grm.

Indications and Contra-indications.—(1) Urea stibamine is indicated in all cases of kala-azar. It has been used in cases complicated with bronchitis or dysentery without any untoward results. In cases of leishmania dysentery it cures the condition. If, however, dysentery develops during treatment, then it is desirable to stop the injection for a time or give it in smaller doses. (2) In cases with marked nephritis and œdema, begin with smaller doses. If the œdema increases, give the doses at longer intervals than before. (3) Very advanced cases should be treated, beginning with small doses which should be slowly increased.

Advantages of Treatment with Urea Stibamine.—(1) The short course occupying only two to three weeks for a complete cure. (2) The appearance of early and marked leucocytosis, rapid disappearance of the fever, of splenic and hepatic enlargements and of anæmia, œdema and cachexia, in fact, of all the symptoms of the disease, and reversion to normal state of health. (3) The absence of symptoms of intolerance after its administration. (4) It is most valuable in the treatment of relapses, or in the cases resistant to sodium antimonyl tartrate or tartar emetic. (5) Observations have shown that early cases are cured after four or five injections, and sometimes even after fewer injections.

In a series of twenty-eight successive cases in the wards of the Calcutta Medical College Hospitals, under the different physicians, the temperature came down to normal in fourteen after the first injection of urea stibamine, in six after the second injection and in six after the third injection, i.e., in 50 per cent. of the cases the temperature came down to normal after the first injection.

In a combined series of 194 successive cases observed by the different physicians in the indoor and outdoor departments in the above hospitals, and by Shortt and Sen, as well as by Greig and Kundu, in the Kalar-azar Research Hospital at the Pasteur Institute, Shillong, the total average amount of urea stibamine required for cure was 1·71 grm. This includes the average actual amount of urea stibamine used, the average sterilizing dose as estimated by blood-culture being less than this.

In the same series as above, the average number of injections per patient required for cure was 8·25, and the average number of days was 16·6.

As already noted, cases of cure with much smaller amounts of urea stibamine and with much fewer injections and in much shorter periods, have been observed, as will be seen from the following table :—

TABLE SHOWING A NUMBER OF CASES CURED BY LESS THAN ONE GRAMME OF UREA STIBAMINE.

	Cal. Med. Coll. Hospital Barnardo, McCay, Author					Pasteur Inst., Shillong Shortt, Greig and Kundu		Author and Maity			
Duration of illness in days	60	90	10	21	21	240	17 mths.	90	—	—	-
Amount grm. after which cult. neg.	0·55	0·65	0·55	0·9	0·4	—	0·95	0·7	0·7	0·5	0·6
Total amount grm. given	0·55	0·8	0·7	0·9	0·55	0·65	0·95	0·7	0·7	0·7	0·6
Period of treatment in days	15	14	10	22	14	6	7	5	6	9	5

In the cases recorded above the proof of cure was based on repeated blood-cultures and keeping the patient under observation, which in some cases in the Calcutta Medical College Hospitals extended from three to five months after completion of treatment.

In a combined series of 325 published cases, 98·47 per cent. of the cases were cured. One of the cases died of extreme asthenia, being admitted at the age of 65 in a moribund condition. In 298 of these cases, proof of cure was microscopic and cultural examinations and disappearance of symptoms, and in twenty-seven cases proof of cure was clinical disappearance of the symptoms and subsequent observation of the cases. One case was resistant.

Other Aromatic Therapeutic Antimonials.

Stibamine is the name given by the author to p-amino-phenyl-stibinate of sodium (*Journ. Trop. Med. Hyg.*, 1921). It is the antimony analogue of p-amino-phenyl-arsenate of sodium (= atoxyl or soamin). It has been prepared in the author's research laboratory. The pure compound is fairly stable and its solution is neutral. The author was the first to introduce the compound in the treatment of kala-azar, and reported a few cases showing its curative value. One case was cured with 1·2 grm. of the compound. It is slightly more toxic than urea stibamine.

Metachlor-acetyl-p-amino-stibinate of sodium was prepared in the author's research laboratory and was named by him chloro-stibacetin. It has been supplied by von Heyden under name of von Heyden "471," and subsequently under the name of stibosan. The total average amount required for cure with chloro-stibacetin was 2·75 grm., and the average number of injections was fifteen. Napier seems to be the only observer who has given an extensive trial to the compound. In his series the average number of injections for cure was 13·3, and mean number of injections prior to the cessation of fever was 5·6, and the mean total dose was 2·78 grm., and the death-rate in his series was 10·6 per cent. In thirteen cases treated with the compound by Greig and Kundu, the average amount required for cure was 2·0 grm. and the percentage of uncured cases was fifteen, one case proving resistant.

Stibiglycine-amide is the name given by the author to N-phenyl-glycine-amide-p-stibinate of sodium. The author has published records of his observations on eight cases showing the value of this compound in the treatment of kala-azar, all the cases being cured (*Indian Journal of Medicine* and *Calcutta Medical Journal*, June, 1926).

The Glucose Compounds.—It has been observed by the author that stibamine, urea stibamine, and other allied compounds have the property of combining with glucose in molecular proportion. Weight for weight the resulting compound for urea stibamine is weaker in its therapeutic properties in proportion to its antimony content. Glucose stibamine has been prepared in the author's research laboratory by the combination of glucose with stibamine in molecular proportions. It has been put on the market under the name of neostam (B.W. and Co.). In Napier's series of eleven cases treated with neostam, the mean

number of injections was 13·8 and the mean total dose was 2·58; the mean number of injections prior to the cessation of fever was 5·4. There was one death in this series of eleven cases. Greig and Kundu tried this compound in two cases only, and therefore no conclusion can be drawn from these two cases.

Para-amino-phenyl-stibinic acid in combination with urea and glucose. In Napier's series of fifty-two cases treated with p-amino-phenyl-stibinic acid in combination with urea and glucose, there were two deaths and two resistant cases not yielding to the drug. This gives a percentage of about eight for uncured cases in his series.

Von Heyden "693" has been used by Napier in the treatment of kala-azar.

The Therapeutic Value of the Different Aromatic Antimonials Compared.

The comparative value of the aromatic antimonials in the treatment of the disease depends upon their toxicity and therapeutic effects following their administration. These again depend upon their chemical configuration and their physico-chemical properties.

(I) *The Aromatic Antimonials of the Stibino-benzene Group.*

Antimonials of the stibino-benzene type have not yet come into use in the treatment of human diseases, though they have been used with indefinite results in the case of certain diseases of animals.

Trivalent aromatic antimonials of the type of salvarsan or neo-salvarsan will probably be in future the further advance in the antimony treatment of kala-azar.

(II) *The Aromatic Antimonials derived from p-stibanilic acid (p-amino-phenyl-stibinic acid).*

Frankel is not justified in stating that changes in the molecular structure of antimony compounds do not bring about an increase of their therapeutic properties. Urea stibamine is just as useful in the treatment of kala-azar as atoxyl in diseases for which it has been recommended. Taking the published cases of different observers, under different conditions and in different places, it is obvious that of all the aromatic antimonials, the most extended trial has been given to urea stibamine. On the other hand, observations upon the other aromatic antimonials are mostly limited to observations of single individuals.

It will be seen from the above that urea stibamine is the most powerful of all the aromatic antimonials so far used in the treatment of kala-azar. Regarding stibamine and N-phenyl-glycine-amide-p-stibinate of sodium, no comparison can be made at present with the other aromatic antimonials, as neither of them has yet been given an extended trial in the treatment of the disease.

On theoretical grounds, N-phenyl-glycine-amide-p-stibinate of sodium, or stib-glycine-amide as the author has named it, should be of very marked therapeutic value, as it contains the group NH_2CO and is allied to tryparsamide.

Napier's observations lead to the conclusion that the chloro-stib-acetin (= stibosan, von Heyden) is weaker in its therapeutic effects when compared with urea stibamine.

The therapeutic value of the glucose compounds as compared with the compounds from which they are derived is proportional to their antimony content, and the same conclusion is arrived at on theoretical considerations.

The author has not yet tested the value of von Heyden "693." It is stated to be p-amino-phenyl-stibinic acid in combination with an amine, but this gives no idea about its chemical composition.

The antimony content of some of the aromatic antimonials is given below :--

Stibamine	42·10 per cent.
Urea stibamine	36·05 ,,
Chloro-stib-acetin (= stibosan, von Heyden "471") ...	33·3 ,,
Glucose-stibamine	25·8 ,,
Glucose-urea stibamine	23·8 ,,
N-phenyl-glycine-amide-p-stibinate of sodium	29·3 ,,

It may be stated that, generally speaking, the therapeutic value of the aromatic antimonials derived from p-amino-phenyl-stibinic acid is proportional to their antimony content.

As stated before, urea stibamine has been observed to manifest no deterioration or other changes, either in physical and chemical characters or in therapeutic properties, if kept in sealed ampoules under ordinary conditions. It has been claimed that antimony compounds which can be stored in ordinary stoppered bottles and weighed out when required are the most useful for general purposes. In the author's opinion such compounds must have more or less the same stability as the antimonates and, therefore, there is less chance of the production of the reactive — Sb = O in the tissues after their administration which, the author considers, is responsible for the beneficial results following the administration of an antimony compound. This explains why antimonates in which antimony exists in the pentavalent form are of very little value in therapeutics, as they are very stable and quickly excreted unchanged after this administration. The same fact also holds good in the case of arsenic.

As regards toxicity, the author has observed that the toxicity of compounds obtained from para-stibanilic acid is proportional to their antimony content. His observations are different from those of Napier, who found urea stibamine and its glucose derivative equally toxic. This latter observation is somewhat significant, as this would mean that the antimony content of urea stibamine is one half times less toxic than its glucose derivative.

COMPARISON OF THE THERAPEUTIC VALUE OF THE VARIOUS ANTIMONIAL PREPARATIONS AND THE MODES OF ADMINISTRATION IN THE TREATMENT OF KALA-AZAR.

Until recently the antimonyl tartrates were most commonly used in the treatment of kala-azar. Since the introduction of urea stibamine by the writer, the antimonyl tartrates are being quickly replaced by this new com-

pound. As stated before, its advantages include the short course of treatment, occupying only two or three weeks or less for a complete cure in many cases, and the rapidity with which the symptoms of the disease disappear. Cases that resist tartar emetic or sodium antimonyl tartrate quickly yield to treatment with urea stibamine.

Antimonious oxide suspended in solution of sugar and gum arabic, or in 30 per cent. emulsion in oil (trixidine), has been found to be of therapeutic value in the treatment of trypanosome-infected experimental animals. Yorke and Blackmore have also used trixidine in oily suspension intramuscularly with beneficial results. Kolle, Hartoch, Rothermundt and Schürmann consider that the formation of a deposit of an insoluble slowly absorbable compound of antimony, such as antimony trioxide, acts prophylactically against trypanosome infection. The principle of the employment of insoluble organic compounds of antimony, either in ointment form or through the formation of intramuscular depots, constitutes what the authors designate *therapia mite curans*, as contrasted with *therapia magna sterilans*. Rogers used it in kala-azar. In the author's experience it is weak in its therapeutic properties in kala-azar. It may perhaps be used in kala-azar in resistant cases, along with intravenous medication, and evidently an intramuscular injection of such a compound is likely to be more useful than substances of the nature of T.C.C.O.

Plimmer and Bateman opened up a new vista when they showed that antimony in metallic form was more effective in trypanosomiasis than its soluble salts. The metal can be prepared in a state of very fine subdivision. It was first used intramuscularly in various media, such as Lambkin's medium. The intramuscular injection is very painful. It was subsequently recommended to be given intravenously, and the results obtained surpassed in its therapeutic effects any of the preparations of antimony that were used before. Ranken found that it never gave rise to any blocking of the capillaries, being quickly absorbed by the leucocytes.

The author's observations have proved the superiority of metallic antimony over the antimonyl tartrates. In some cases in which the soluble salts, such as potassium antimonyl tartrate and sodium antimonyl tartrate, did not show any improvement in the blood condition or the temperature of the patient after several injections, metallic antimony brought about complete cure. Such cases belong to the refractory types of the disease, but are not uncommonly met with.

In addition, the number of injections required for a course of treatment with metallic antimony is much smaller than those of the tartrates. Three or four injections frequently cure the patient, though in one or two cases the injections given were as many as eight or nine. Even then the number of injections is less than what is generally required in the case of the antimonyl tartrates.

The only objection is, no doubt, the complicated technique of the operation of injection, which is a serious obstacle to mass treatment of the disease.

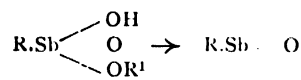
In previous works on kala-azar the author held the view that metallic antimony was the most powerful of all the leishmanocides, and there is no doubt that this was so till his introduction of the aromatic antimonials in the treatment of kala-azar. His present view is that, apart from the complicated method of its administration, it is one of the most powerful drugs in the treatment of kala-azar. It should be tried in those resistant cases in which other antimonials may fail.

In studying trypanosomiasis, Ehrlich thought that the trypanosomes assimilated the organic derivatives of arsenic only when the arsenic was present in the trivalent and not in the pentavalent form. Similarly, the experiments of Kolle, Hartoch, Rothermundt and Schürmann have shown that compounds containing pentavalent antimony were not organotropic, except in large doses, and were at the same time slightly parasitotropic. Preparations containing trivalent antimony were, as a rule, exceedingly toxic to the organism, and at the same time it was also shown by these observers that for antimony compounds, soluble or insoluble, organic or inorganic, to be of therapeutic value in trypanosomiasis, antimony must be in the trivalent form.

The more recent researches of the author and others that have followed him have, however, proved that the aromatic pentavalent antimonials are much more potent in the treatment of kala-azar than the trivalent antimonyl tartrates.

As stated before, antimonials of the stibino-benzene type have not yet come into use in the treatment of human diseases, and that trivalent aromatic antimonials of this type will probably be in future the next advance in the antimony treatment of kala-azar.

By studying the excretion of antimony in man after intravenous injection of the aromatic antimonials of the type of urea stibamine, and also of the antimonyl tartrates, the author has been able to explain the superiority of the former over the latter. He has observed that in the case of tartar emetic the curve of excretion is one slowly converging to the base line. The following extracts are made from his paper in the *Indian Journal of Medical Research*, January, 1924: "By studying the excretion of antimony we have come to the conclusion that antimonials of the type of urea stibamine are converted within the body into trivalent oxide of antimony, similar to what happens in the case of organic pentavalent arsenicals, before they are capable of exhibiting organotropic and parasitotropic properties. This may be illustrated graphically in the following way :—



"It is possible that – Sb = O is more reactive and more powerful in its parasiticidal properties when an organic antimonial is changed within the body into one containing it than when it exists in the antimonyl tartrates. We may for the present assume that the parasiticidal properties of antimony compounds depend upon the radical – Sb = O, similar to what exists

in the case of arsenicals, in which the parasiticidal properties depend upon – As = O." The amount of antimony excreted in the urine during the first twenty-four hours after intravenous injection of tartar emetic is about 6 per cent. of the amount injected. The amount of antimony excreted in the urine during the first twenty-four hours after intravenous injection of urea stibamine is 30 to 40 per cent. of the amount injected. The excretion of antimony after intravenous injection of a pentavalent organic antimonial follows a curve, the first portion of which, representing the excretion during the first twenty-four hours, is abrupt, and the second portion follows a course similar to that found in the case of tartar emetic. It is probable that a pentavalent organic antimonial is converted in the body into a trivalent antimonial, and that as long as it exists in the body in the pentavalent form its rate of excretion is much quicker than when it is converted into the trivalent form. During the latter stage the curve of excretion is similar to that of tartar emetic, in which antimony exists in the trivalent form. Since a great portion of antimony present in an aromatic pentavalent antimonial (urea stibamine) is quickly eliminated, the chances of toxic action of the compound are much less than that of an antimonyl tartrate. In the process of conversion of an aromatic pentavalent antimonial in the body into a compound containing trivalent antimony, a reactive – Sb = O is formed, which is probably responsible for the remarkably beneficial results observed in the treatment of leishmaniasis by the use of urea stibamine. The low toxicity of urea stibamine, the fact that no intolerance towards the drug is likely to take place on account of the quick elimination of a large proportion of it, the fact that it is perhaps converted in the body into a trivalent antimonial containing a reactive – Sb – O radical, and the fact that its therapeutic value is very great, make urea stibamine the best of all the antimonial compounds that have so far been discovered for the treatment of leishmaniasis and other diseases in which antimony is indicated.

As has been proved before, clinically speaking the same conclusion has also been arrived at. We would close the chapter by quoting the remarks of Shortt and Sen, which they made in 1925: "We consider that the value of urea stibamine has been established as the most efficient drug at present in use in the treatment of Indian kala-azar." This statement, according to the present writer, remains equally true to-day. To this may be added the remarks of Dodds Price: "I am of opinion that urea stibamine is a most valuable remedy in the resistant types of the disease, and I strongly urge that it should be resorted to if, after a few injections of sodium antimony tartrate, a patient does not show marked improvement." The author would only add that metallic antimony should be resorted to in those very few cases which may be resistant to urea stibamine, and which perhaps do not go beyond 3 per 1,000, or less.

Up to now the intravenous method of administration is the most effective form of treatment. The late Sir Patrick Manson once wrote to me as follows: "Go on in your efforts to get an antimony compound that can be used as an intramuscular injection, or, better still, as a drug that can be

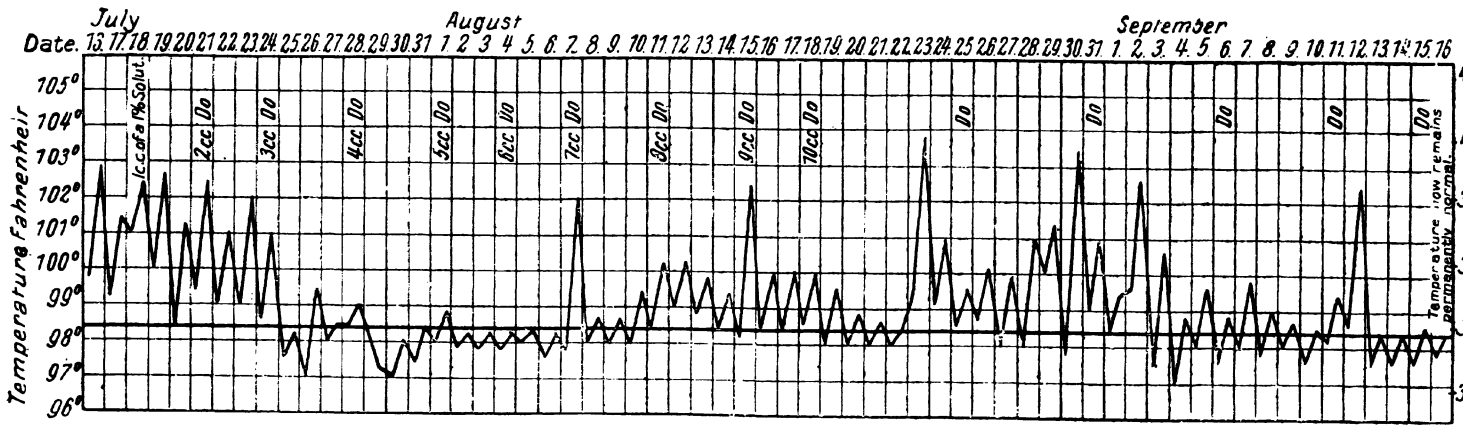


FIG. 34.—Temperature chart of a case cured by intravenous injections of bismuth tart. solubilis.

administered by the mouth." The therapeutic value of an antimonial depends upon its concentration in the tissues after administration. Unfortunately, ordinary antimonials cannot be administered orally, intramuscularly, or per rectum, in such doses as to bring about this concentration, without at the same time giving rise to local distressing symptoms. The new amino-antimonyl tartrates containing radicals, possessing anæsthetic properties, and to which the author has already referred, may be worth trial by these routes. Ointment of metallic antimony in a state of finest subdivision may be more easily absorbed and less irritating than that made with ordinary metallic antimony.

"BAYER 205."

In a few cases "Bayer 205" has been used by the author, but the results have been unsatisfactory. Others, with the exception of Yorke and Arcoleo, have had the same experience. Yorke reports one case which was apparently benefited by this compound.

BISMUTH.

In a limited number of cases the author has used bismuth compounds in the treatment of the disease. Soluble bismuth tartrate, bismuth in the form of precipitated metallic bismuth suspended in isotonic salt solution (neo-trepol), or tartro-bismuthate of potassium and sodium (trepol) may be employed for the purpose. The dose of soluble bismuth tartrate is 1 c.c. to 10 c.c. of a 1 per cent. aqueous solution given twice a week as an intravenous injection. In a few cases the patient made a complete recovery. It is as yet too early to compare the relative curative values of antimony and bismuth in the treatment of kala-azar.

Fig. 34 shows the temperature chart of a patient cured with intravenous injections of bismuth.

Ancillary Treatment.

In addition to specific treatment with antimony, symptomatic treatment for complications has to be given when required (see Chapter on treatment of Complications). Generally speaking, no ancillary treatment is nowadays needed. In some cases in which the leucocytes do not increase with proper rapidity, the various leucocyte-increasing methods of treatment may be adopted, but generally they are of no avail. A short account of the methods that were adopted in the pre-antimony days is given by the author.

Attempts have been made from time to time to treat the disease by local irritation with the idea of bringing about leucocytosis. The use of blisters over the spleen, or the introduction of a seton into the muscles of the arms or the legs, was advocated long ago in the treatment of enlargement of the spleen. In some cases the use of the seton was followed by a septic infection with an excessive pus formation, resulting in apparent diminution of the spleen and amelioration of the symptoms or complete cure of the disease. In

other cases, the treatment was not followed by any good results. In some places the patients used to insert a seton consisting of a piece of dirty string through a loop of skin over the spleen, thus keeping up a constant irritation or suppuration. In Bengal, sometimes, a piece of dirty wood is inserted into the muscles of the arm or leg or over the splenic region. Sometimes the skin is scarified over the spleen and the leaf of a tree of the ficus order rubbed in, followed by the application of a powder made by burning the core of the jack fruit.

Muir states that injection of turpentine increases the polynuclear leucocytes of the blood. He advocates a solution of one part of turpentine, camphor and creosote, and two half-parts of olive oil (T.C.C.O.), five to fifteen minims of which are injected into the muscles on both sides of the body, the latissimus dorsi or gluteus being most suitable.

These injections are very painful and are not always followed by leucocytosis, and do not seem to influence the course of the disease. Sometimes blisters have been caused over the splenic region by the application of liq. epispasticus, and ulcers produced thereby have been kept up by application of ung. sabinæ. Sometimes, again, after cutting into the skin and subcutaneous tissue, aseptic glass balls have been introduced into the cuts ; the deep ulcers produced thereby have been kept up by the retention of these balls. Though it has been possible to bring about a temporary leucocytosis, the effect is never permanent and the ulcers tend to heal up. The effects, on the whole, are unsatisfactory. The author has never seen a single case cured simply by the injection of turpentine.

In cases of experimental trypanosomiasis Ciuca has obtained better results by the combination of atoxyl with production of abscess of fixation by injecting 0·5 to 1 c.c. of essence of terebene as high as possible on the external surface of the thigh in cases of experimental trypanosomiasis, than with atoxyl alone. He thinks that the abscess of fixation produces trypanotoxyl more quickly and efficiently.

It has been claimed that in cases which have hard fibrous spleen, one or two injections of T.C.C.O. will bring about a considerable softening and diminution in the size of the spleen. This, however, has never been observed by the author, and it is difficult to understand how the fibrous tissue of a fibrous spleen can be softened by its administration. As stated before, it may be advantageous to give an injection of trixidine in some cases to try to bring about inflammatory leucocytosis, and to combine with it the beneficial effects of antimony.

Attempts were made by the author to bring about leucocytosis in kala-azar by oral, subcutaneous or intravenous administration of hetol. No permanent leucocytosis has been obtained by this method. In some cases a temporary increase of leucocytes was obtained, but this quickly disappeared. In one case the patient was given more than 100 injections of hetol.

In one case the patient was given 25 injections of nuclein in 20-minim doses on successive days, but no improvement followed in the leucocyte count of the blood. In another case the patient was treated with hetol (gr. iij) and

nuclein (minims xx) given intravenously on alternate days for nearly two months. There was slight improvement in the general condition of the patient and apyrexia existed for some time. Yeast has been tried internally without any benefit.

Rogers used 0·1 to 0·4 grm. of sodium nucleate subcutaneously. The effect on the leucocytes was very disappointing, frequently no increase of the leucocytes taking place, while in no case did any slight rise which ensued persist for any length of time. Phylacogens have been tried by Rogers as well as by the author without any good results.

Leucocyte extract has not yet been used for bringing about leucocytosis in kala-azar, and may be probably worth a trial to bring about increase of leucocytes. Leucocyte extract was tried in trypanosomiasis by Alexander. This was prepared by injecting Mellin's food into the pleural cavities of rabbits. The rabbits were killed and the accumulation of leucocytes was taken from the pleural cavities and extracted with sterilized distilled water. One injection produced a very great increase of leucocytes in the blood on the following day. Infantile kala-azar was treated by Nicolle and Cassuto by hemoplase (extract of red corpuscles) in doses of 4 c.c. along with atoxyl. There was a great improvement in the general condition, but the patient subsequently died during a violent attack of dyspnœa. These methods of treatment have not been tried in Indian kala-azar.

In a few cases Coley's fluid was used by the author. There was slight increase of the leucocytes in some cases, but the effect was not permanent.

Rogers made an extensive use of vaccines in the treatment of kala-azar. A number of cases were treated by repeated subcutaneous injections of dead staphylococci. In almost all of them there was marked increase of the leucocytes. The improvement was temporary in nature, and a time came when the injections of the vaccine failed to further increase the leucocytes or improve the general condition. Rogers further tried subcutaneous and intravenous injections of sensitized living staphylococcus vaccine, and claimed to have obtained satisfactory results by combining them with spleen tabloids and alkalies.

Bassett-Smith described a case of kala-azar treated with some benefit with intramuscular injection of soamin (3 to 5 gr.) twice a week, together with some injections of an autogenous vaccine prepared from cultures of the patient's own flagellates.

Row got favourable results in the allied disease of oriental sore by injection of a vaccine derived from cultures of the *Leishmania* of oriental sore.

The high fever may be treated with baths, and anæmia by injections of soamin or sodium cacodylate. Along with this, iron may be administered. Generally speaking, the anæmia disappears during treatment with antimony.

According to observations made in India, the effects that may follow a change of climate without any specific treatment are slight, and only a very few cases are benefited by this. In Italy Gabbi also tried the effect of cures in the mountains. In a few cases of infantile kala-azar the blood improved and the fever subsided. The improvement, however, was only temporary.

SYMPTOMATIC TREATMENT AND TREATMENT OF COMPLICATIONS

Dysentery.—Dysenteric symptoms closely allied to those of true dysentery may appear during the course of kala-azar. Such cases of *Leishmania* dysentery should be treated with antimony. In many cases, however, dysentery appears as a terminal infection, being generally of the bacillary variety, although sometimes amœbic dysentery may complicate the disease. In most cases dysentery contra-indicates treatment with antimony, and it is difficult to make out whether the dysentery is due to *Leishmania* or not. Bacillary cases frequently terminate fatally. Septic dysentery is almost always fatal. Cases which are of the amœbic variety should be treated with emetine. Such cases are rare, and emetine frequently fails. In some cases rectal injection of argyrol (10 per cent.) after irrigation of the rectum with normal saline or boric acid lotion may be helpful. Dover's powder with pulv. cretæ aromaticus and tannigen may also be tried. Salol or benzonaphthol or dimol may also be tried. The diet should be most carefully regulated in cases complicated by dysentery.

Septic Infections.—Cancrum oris is a frequent terminal complication of the disease. In some cases antimonial treatment leads very quickly to a cure of the cancrum oris. In other cases the treatment does not seem to influence the course of the disease and the patient dies, in spite of the antimonial treatment. For local treatment of cancrum oris, the part should be swabbed with a mixture of trichloracetic acid and glycerine (1 in 8). Cotton-wool soaked with the above may be kept in contact with the affected part and changed every twelve hours. In this way the slough may separate. In some cases the parts have been soaked with colloidal silver with good results. Gargles, consisting of eusol or other antiseptics, such as electrolytic chlorine, flavin, acriflavin or dichloramin may be used. Irrigation with lysol and keeping the part constantly soaked with the same give satisfactory results. An autogenous vaccine may be used, but generally vaccines have not been found useful in the treatment of cancrum oris in kala-azar.

The occurrence of a definite leucocytosis is a hopeful sign. Nearly all those patients in which it does not occur do not recover.

Other septic conditions, such as mastoid abscesses, otitis media, deep-seated abscesses, &c., should be treated according to general principles. For the treatment of Bright's disease, pneumonia or phthisis complicating kala-azar, books on systematic medicine should be consulted.

Hæmorrhages should be treated with calcium lactate or the other hæmostatics.

Ankylostomiasis should be treated with thymol, β-naphthol, oil of chenopodium or carbon tetrachloride.

Persistent anæmia should be treated with iron and arsenic compounds, such as soamin or sodium cacodylate. Intramuscular injection of blood or a blood transfusion is sometimes found beneficial. In many cases in which there is anæmia and œdema with a dilated heart, the following mixture is recommended :—

℞	Tr. ferri perchlor.	10 min. (0·5 grm.)
	Liq. ammon. acetatis	1 drm. (3·9 grm.)
	Tr. digitalis	5 min. (0·25 grm.)
	Acid. phosphoric. dil.	10 min. (0·5 grm.)
	Glycerine	20 min. (1·0 grm.)
	Water	1 oz. (31·0 grm.)

For a dose, to be given thrice a day.

Cirrhosis of the liver, if it is due to *Leishmania*, may be treated with antimony injections. Ascites due to pressure of cells loaded with *Leishmania* may be cured by antimonial injections.

CHAPTER XV.

RELAPSES AND RESISTANCE TO ANTIMONY TREATMENT.

THE possibility of the existence of antimony-fast *Leishmania* or their development during treatment with antimony has not yet been fully worked out. In the author's experience with the treatment of kala-azar with antimonial preparations, he has met with the following types of cases: (1) Cases which quickly yielded to antimonial treatment; (2) cases that resisted treatment for a considerable period and slowly yielded to treatment; (3) cases that seemed to be extremely or absolutely resistant; (4) cases that got relapses after insufficient or improper treatment with antimony and were either very resistant to subsequent treatment or quickly yielded to it.

From the above, it will be seen that cases vary in their amenability to antimonial treatment and that the possibility of the existence of absolutely antimony-resistant *Leishmania* which may be present from the very beginning or develop during antimonial treatment has to be considered.

Relapses may be divided under the following heads: (1) Relapses due to too early abandonment of treatment; (2) relapses in cases which seemed to have been apparently cured, as shown by marked improvement in the general condition and in the blood-picture and freedom from fever for some months. These latter are very rare.

Leishmania can, therefore, be divided into the following types: (1) Those that are quickly and completely destroyed by antimony; (2) those that are more slowly destroyed by antimony; (3) those that are resistant to antimony from the very beginning; (4) those that become resistant after insufficient antimonial treatment.

The third type is very rare with the recent treatment with urea stibamine. As regards the effects of treatment on relapses, the author has met with the following types of cases: (I) Those that are subsequently and sometimes quickly benefited by further antimonial treatment; (II) those in which further antimonial treatment seems to be very slow in its effects.

As regards (I), the author found originally, before the discovery of urea stibamine, that metallic antimony was the best and the most powerful in the treatment of this type of relapses. More recently he has used urea stibamine with remarkable success. This view has been confirmed by others. Sometimes the substitution of a different antimony compound in place of the one previously used, seems to lead to a cure and may give an impression that one is superior to the other. Thus the substitution of antimonyl sodium tartrate

in place of tartar emetic and *vice versa* leads to recovery in some of these cases of relapse.

As regards (II), it appears to the author that in these cases antimony treatment must be supplemented by a combined therapy. What such a combined treatment should be, is difficult to say.

The treatment of relapses is sometimes a prolonged one, but not always so.

The mechanism of response of *Leishmania* to an antimonial preparation is a very complicated one. While it is universally admitted that urea stibamine brings about sterilization of an infected individual in a much shorter time than tartar emetic or sodium antimonyl tartrate, it is at the same time observed that even with urea stibamine the time required for sterilization is variable. In some cases, this was brought about in seven or less than seven days and in a few cases in thirty-two hours. Though, in some cases, striking results were obtained by the *intensive* method introduced by Brahmachari and Maity, yet there is no doubt that, apart from this, some cases are more quickly amenable to treatment than others. What is the mechanism of this variability ? This constitutes an important line of research. Is it possible that there are in existence different strains of *Leishmania* varying in the action of antimony upon them ?

Levaditi has propounded a general law with reference to all the members of the nitrogen family of elements occupying Group V of Mendelieff's periodic table—arsenic, antimony, vanadium, bismuth, &c. They or their compounds exhibit their parasiticidal properties only after they have been acted upon by the tissues. If fresh extract of liver is added to them, they become actively treponemicidal.

It has been suggested that in the case of bismuth, the action of the cellular extract gives rise to a new compound, "bismoxyl," and it is this which possesses the destructive power against the *Treponema pallidum*. The substance in the extract which has the property of changing bismuth into bismoxyl has been termed "bismogene."

Bismoxyl is a bismuth toxalbumin in all respects similar to those arising from arsenic, vanadium, or antimony.

The present author has suggested that an antimony compound, in order that it may be of therapeutic value, must be converted in the tissues into a compound containing the radical $-Sb = O$ in the reactive stage. Chemically, some of the bismuth compounds contain the radical $-Bi = O$ just as some of the antimony compounds contain the radical $-Sb = O$; and it is very likely that the bismuth toxalbumin also contains the radical $-Bi = O$ in the reactive stage. A corresponding antimony compound which may be called "stiboxyl" is probably formed in the case of antimony. It has been suggested that metallic bismuth, finely subdivided, is more suitable for the production of bismoxyl when administered intramuscularly than in the form of a chemical compound, and the same could have been expected of metallic antimony but for the fact that when injected intramuscularly it gives rise to very severe local irritation, and is therefore unsuitable for intramuscular injection for therapeutic purposes. It has been recently observed by Meleney that in kala-azar, plasmatocyte tissue is developed as a tissue reaction, and

probably, as the present writer has suggested, out of the reticulo-endothelial system. He would further hold that this reticulo-endothelial system gives rise to the production of bismoxyl or stiboxyl, as the case may be. If this were so, then it may be concluded that individual cases will get beneficial results from the use of antimony compounds proportional to the reaction of the reticulo-endothelium system. Two things are necessary, namely, the development of the clasmatocytes and the introduction of an antimony compound with which they can combine for the development of stiboxyl. Herein lies the value of the different antimonials and the superiority of urea stibamine over the other antimony compounds. This also explains why, with the same antimony compound, one individual is cured much more quickly than another after its administration. It is the development of the clasmatocyte cells that one should aim at in the treatment of the resistant cases.

Voegtlin and his co-workers have pointed out that arsenious oxide and its derivatives combine with substances containing a sulphydrile grouping, and that the toxic action of the organic arsenoxides is depressed by the simultaneous injection of excess of sulphydrile compounds. Hopkins has shown that one such sulphydrile compound, reduced glutathione, plays an important part in the hydrolytic oxidation-reduction processes of the living cell. Voegtlin suggests that a combination of the arsenoxides with such groups and consequent suppression of this vital function may explain the toxic and curative actions of the arsenical derivatives, and that a formation by trypanosomes of the sulphydrile compound in excess of its vital need may be the basis of acquired resistance of trypanosomes. The same probably takes place in the cases of *Leishmania*. Investigations in these directions may lead to discovery of methods of preventing the development of antimony-resistant *Leishmania*.

In resistant cases arsenic may anchor some of the *Leishmania* which may not be fixed by antimony alone. A combined therapy of arsenic and antimony may, perhaps, be of value in such cases, just as it is in trypanosomiasis. The combination of substances with different points of attack in the sense of Ehrlich's laws of distribution of medicaments and poisons may enable us to produce combinations of sterilizing substances which may be of value in resistant cases. Whether the combination of three remedies, as has been suggested by Ehrlich and Tsuzuki in trypanosomiasis, or more or less than this number, would constitute the most potent treatment of resistant cases of kala-azar with the least toxicity, will be settled by further investigation.

A combination of antimony, arsenic and bismuth is a possibility for the treatment of such cases. This combination has been recommended by Cushny in the treatment of trypanosomiasis.

Fortunately, under treatment with urea stibamine, relapses and resistant cases are very rare. In cases of relapse after treatment with the antimonyl tartrates, patients should be treated with urea stibamine, and in most cases the results will be satisfactory.

CHAPTER XVI.

DERMAL LEISHMANOID.

Synonyms.—Dermal Leishmaniasis (Brahmachari), Megaw, 1922; Post-Antimonial Dermal Leishmaniasis (Brahmachari), Megaw, 1922; Brahmachari's Dermal Leishmaniasis, Megaw, 1922; Post-kala-azar Dermal Leishmaniasis, Acton and Napier, 1927.

In February, 1922, the author read a paper to the Asiatic Society of Bengal on a New Form of Cutaneous Leishmaniasis which developed in a case of kala-azar cured by antimonial treatment, and which was subsequently described by him in the *Indian Medical Gazette* in April, 1922, under the name of Dermal Leishmanoid. Of all the names suggested this name seems to be the most convenient, on account of its simplicity, and especially in view of the fact that the disease is frequently recognized by the medical profession under this name. A number of such cases have been observed by the author and others since the disease was first described, and it seems to be more common than originally supposed.

The eruptions generally appear about a year after completion of the antimonial treatment of the original disease, as whitish patches on the face, the trunk, and the extremities, associated with papules and nodules, especially marked on the face and sometimes on the scrotum. In the first case, published in the *Indian Medical Gazette*, the blood and scrapings from the lesions were examined for the author by Dr. S. N. Ghose, Bacteriologist, Presidency General Hospital, Calcutta, and the eruptions presented the following characters (see Plate IX):—

(1) On the face there were papillomatous nodules, in some respects resembling small leprotic nodules. Smears from these showed the presence of Leishman-Donovan bodies.

(2) There was a slight erythematous rash on the cheeks and forehead, and similar patches on the extremities, especially the lower.

(3) On the trunk, the upper and lower extremities, there were slightly raised brown patches which were extensively spread over the whole body. Leishman-Donovan bodies were found in the smears from the patches. A few papules were found among these patches.

(4) There was no ulceration or scab formation in any part of the body, no anæsthesia, no loss of knee-jerks, and no thickening of the nerves. No eruptions occurred in the mucous membrane of the mouth and nostrils. The liver and spleen were normal. On examination of the splenic blood by spleen puncture, no Leishman-Donovan bodies were found. There was

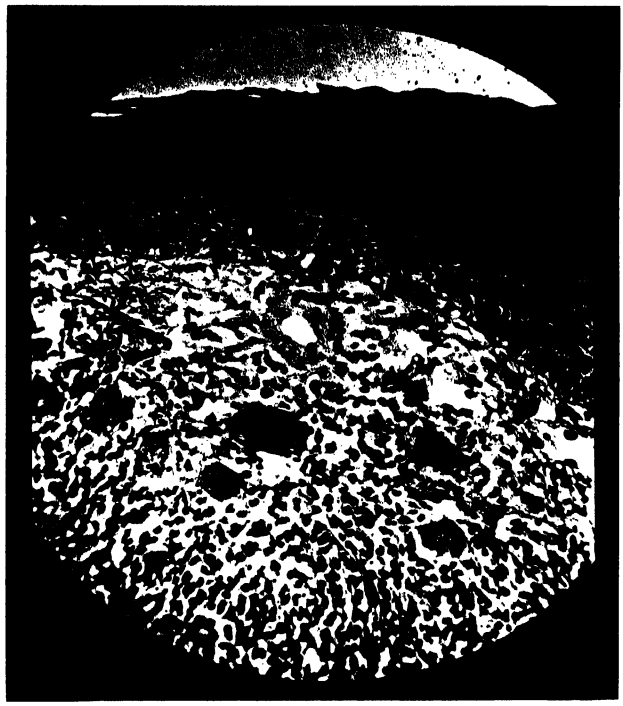


FIG. 35. Section of skin showing giant cells containing Leishman-Donovan bodies.

FIG. 36. Section of skin showing a pigment-carrying cell containing Leishman-Donovan bodies.

FIG. 37. Section of skin showing a highly magnified giant cell containing a large number of Leishman-Donovan bodies.

PLATE VIII.

Dermal Leishmanoid with positive flagellate culture from the peripheral blood in an imperfectly cured case of kala-azar. (Original.)

Face p. 150.

no rise of temperature. The patient complained of no other trouble, except the ugly appearance of the body due to the eruptions.

Result of blood examination showed: Hæmoglobin, 75 per cent.; R.B.C., 4,500,000; W.B.C., 10,000; polymorphonuclears, 62 per cent.; lymphocytes, 24 per cent.; large mononuclears, 6 per cent.; eosinophils, 8 per cent. The blood-picture did not correspond to that of kala-azar. No Leishman-Donovan bodies could be detected in the peripheral blood.

A series of experiments were performed in order to determine whether flagellates developed from the Leishman-Donovan bodies, which had apparently been modified in their virulence by a course of antimonial treatment, and also to determine whether, if monkeys could become infected by transmission, a local or general disease would be produced.

The following are the notes on the cultural and inoculation experiments kindly made for me by Major Knowles, in one of the cases:—

(1) One of the nodules of the right arm was pricked, and the blood which oozed out was inoculated into N N N medium and incubated at 22°C. Flagellated bodies were found after twelve days; these were indistinguishable from those of *Leishmania donovani* (Plate X, fig. 2).

(2) The culture and smears from the peripheral blood of the patient and examination of the splenic blood all gave negative results.

(3) A monkey (*Macacus rhesus*) was inoculated in both eyebrows by embedding bits of granulomatous nodules into pockets cut in them. After one and a half months marked granulomatous growths were observed over the sites of inoculation in both eyebrows. Small secondary nodules were also observed at the outer and inner canthuses of the eyes. One of the nodules at the original site of inoculation was incised and smears were made from it. A fair number of Leishman-Donovan bodies were present, most of which were intracorpuscular and a few only being extracorpuscular (Plate X, fig. 3).

(4) No ulceration was observed over the nodules after two and a half months, the raw surface left after incising one of the nodules having healed up.

(5) Blood examination and cultures made from the peripheral blood of the monkey gave negative results a month and a half after inoculation. The smears from the liver of the monkey and cultures from the same organ on N N N medium gave negative results, a month and a half after inoculation.

HISTO-PATHOLOGY (MODIFIED FROM SHORTT).

The following is the description of the histo-pathology of the nodules cut in a direction perpendicular to the surface of the skin.

The lesions appear to be specific infective granuloma.

APPEARANCES UNDER A LOW POWER (fig. 38, and Plate X, fig. 4).

The first glance shows that there is a profound alteration in the structure of the superficial tissues. This alteration implicates both the epidermis and the *cutis vera*, and these will be considered separately.

Epidermis.—The structural modification here consists in a uniform attenuation of the epidermis affecting all the recognized layers, but most

evident in the *rete mucosum* on account of the relative diminution in number and length of the finger-like processes usually associated with this layer in normal skin. While the thickness of the epidermis is thus greatly reduced, there is, at the same time, no tendency for this process to proceed to the extreme degree of ulceration.

Cutis vera.—It is in this region that the most striking changes are manifest. The normal condition of dense connective tissue, merging gradually into the more open subcutaneous tissue, is entirely replaced by what appears to be a

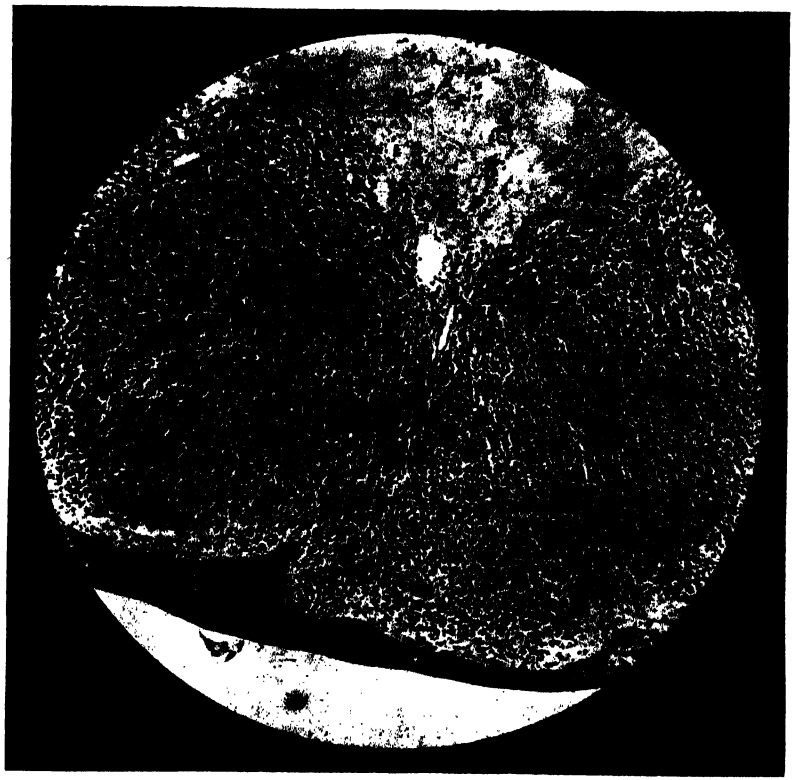


FIG. 38.—Section of skin in dermal leishmanoid. After Shortt, in Shortt and Brahmachari's paper. (Low magnification.)
(From the *Indian Journal of Medical Research*, January, 1925.)

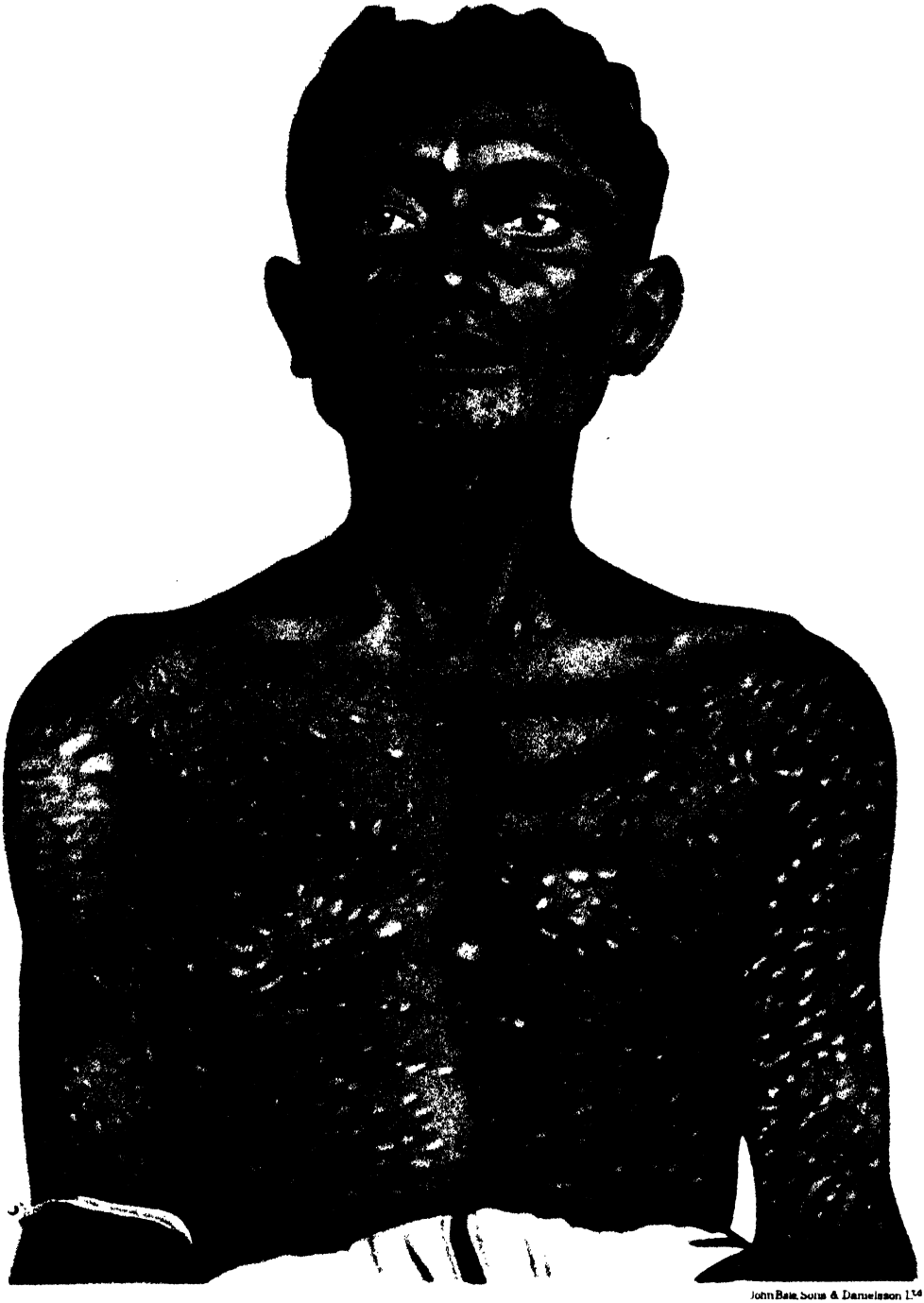
dense infiltration of cells, forming a deep layer sharply differentiated from the underlying open structure of the subcutaneous tissue. This cellular layer averages in depth about nine times the depth of the epidermis.

APPEARANCES UNDER A HIGH POWER (figs. 35, 36, 37 and 39, and Plate X, fig. 5).

Epidermis.—Beyond a diminution in the thickness of the layers the minute structure is unaltered.

Cutis vera.—This appears to be composed of a very primitive connective

PLATE IX.



John Bale, Sons & Danielsson Ltd

Dermal leishmanoid in a cured case of kala-azar.

FIG. 1. *Leishmania donovani* in a smear from a papule. Some few are free and extracorpuscular, but most are inside leucocytes and endothelial cells. (Eye-piece 4. Objective $\frac{1}{12}$ oil immersion.)

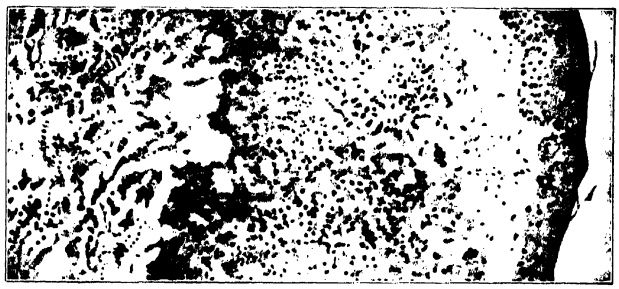


FIG. 4.– Section through a skin papule of a patient. Round-celled infiltration with fibroblasts and thinning of the epidermis. (Low magnification. Eye-piece 4. Objective Zeiss A.)

FIG. 5. –The same section as in fig. 4, showing a network of newly-formed capillaries and thickening of the capillary wall. (Eye-piece 4. Objective Zeiss D.)

FIG. 2.—Flagellate culture on N N N medium twelve days after incubation at 22° C. (Eye-piece 4. Objective $\frac{1}{12}$ oil immersion.)

FIG. 3. *Leishmania donovani* in a smear from a nodule of the eyelid of a monkey. (Eye-piece 4. Objective $\frac{1}{12}$ oil immersion.)

Once the primitive cell has become definitely differentiated into a stroma cell or into capillary endothelium, it seems to lose its power of active phagocytosis. The parasitized cells are mainly situated close under the epidermal layer and become fewer in number in the deeper parts of the tissue. Many of these cells, besides containing parasites, enclose abundant pigment granules. The behaviour of the pigment cells of the epidermis in the affected portions of the skin is very interesting. They appear to be infected with *Leishmania* from the very earliest stages of the skin lesions. Some of them become very much swollen (see fig. 36), due to the presence of *Leishmania*, others probably take on the character of spindle cells and appear to be converted into fibroblasts. The parasitized pigment cells tend to migrate from the lowest layer of the epidermis into the granulation tissue.

The areas of de-pigmentation noticed over the nodules contain much fewer pigment cells than in the adjacent structure of the healthy skin, and this de-pigmentation is partly to be explained by the migration of the parasitized pigment cells. The connective tissue underlying the *cutis vera* is quite normal in appearance.

Here and there the endothelium of the large capillaries is hypertrophied and the wall thickened. In some cases this occurs to such an extent that the capillaries are almost obliterated. On the wall of the thickened capillaries and in their endothelial lining Leishman-Donovan bodies are seen. The skin over the papillomatous nodules is slightly pigmented and the stratum cornium is very thin.

On the whole the pathological changes in the skin are very similar to those present in a non-ulcerated oriental sore (see Plate X, figs. 4 and 5), and no break of continuity of the epidermis is observed over the nodules as shown by ulceration.

The Leishman-Donovan bodies are best seen in smears from the scrapings from the granulomatous nodules. In these smears they apparently appear as free parasites. On careful examination they are found inside large mononuclear leucocytes, and rarely inside the polymorphonuclears. More commonly, they are found inside other large cells, which possibly originate from the endothelium (Plate X, fig. 1). Parasites are also found in the brown patches.

After attention was drawn to the disease by the author, cases have been studied by Acton. An early de-pigmented stage, a later nodular stage and a xanthoma type of the disease in which there is a tendency towards fibrous tissue formation and constriction of the venules and subsequent dilatation have been described (Acton and Napier). It is stated by them that the nodules appear in the place of de-pigmented patches.

In the author's experience the disease first shows itself as erythematous patches over the skin before any de-pigmentation or nodule appears in the skin. Besides nodules may appear in places where there have been no de-pigmented patches, and there may be extensive areas of de-pigmentation lasting for several months without any tendency towards formation of nodules in their place. Further, the tendency towards fibrous tissue formation is

Dermal leishmanoid in a cured case of kala-azar. (From a case of the author in the *Indian Journal of Medical Research*, January, 1925.)

slight, just as in the case of the spleen in kala-azar. When the cases recover the skin assumes its normal appearance without any sign pointing to the presence of increased fibrous tissue formation.

Besides the above, the following types of infection of skin with *Leishmania donovani* have been observed.

(1) Dermal lesion in imperfectly cured cases of kala-azar, in which positive flagellate culture was obtained from the peripheral blood at the time of observation (see Plate VIII).

In the case shown in the Plate (VIII), with positive flagellate culture from the peripheral blood, the patient was at first treated with sodium antimonyl tartrate, and when the nodular eruptions appeared on the body, he was treated with stibosan without any beneficial results. When he came under the observation of the author and Dr. A. M. Dutt, of the Calcutta Medical College, he was still suffering from kala-azar with the skin eruptions. He was completely cured of kala-azar after treatment with urea stibamine, the skin lesions slowly but subsequently completely yielding to a prolonged treatment with the compound (*Indian Journal of Medicine* and *Calcutta Medical Journal*, February, 1927). This is the same case as Case 6 described in Acton and Napier's paper on post-kala-azar dermal leishmaniasis (1927).

(2) Cases of dermal lesion showing *Leishmania donovani* in the skin in which there was no previous history of kala-azar and observed in endemic areas of the disease (Annual Report of the Calcutta School of Tropical Medicine, 1925, and *Indian Journal of Medicine*, September 1926).

It is impossible to say with certainty whether the skin lesions in this type are due to infection by *Leishmania donovani* or *Leishmania tropica*, though the fact that the patients came from places where *Leishmania tropica* is unknown, and *Leishmania donovani* infections are very common, goes strongly in favour of their being due to infection by the latter.

(3) Untreated cases of kala-azar, in which sometimes Leishman-Donovan bodies may be found in minute papules in the skin, as was observed by Christophers.

The skin eruptions under (2) and (3) quickly disappear under antimony treatment.

We like to extend the term dermal leishmanoid to signify skin lesions due to *L. donovani* in cases which manifest no signs of internal leishmaniasis either as a result of previous treatment with antimony or without it. In those cases in which the skin lesions appear about a year after the patient has been cured of kala-azar, the leishmania in the skin are very resistant to subsequent antimony treatment.

Cases of dermal leishmanoid are not so uncommon as was originally supposed by the author. Skin lesions due to *L. donovani* in imperfectly cured cases of kala-azar may perhaps be also termed dermal leishmanoid. (See Addendum for further observations on peripheral lesions due to *L. donovani*.)

CHAPTER XVII.

PROPHYLAXIS.

(A) India.

Rogers was of opinion that the disease was caused by a house-infection, and he recommended that all patients from houses where the infection had first occurred should be removed to a place at some distance from the infected area.

The Garos, in the early stages of the Assam epidemic, learnt that the best way to escape being more than decimated by the scourge was to move from the infected villages to a new site. Dodds Price, suspecting the disease to be infectious, placed in a tea garden a number of freshly imported coolies in newly-built lines of houses, while the remainder had to be accommodated in infected lines for want of room. It was ascertained by him that although none of those in the new lines had suffered from kala-azar during two years they had been in this badly-infected tea garden, yet no less than 8 per cent. or 16 per cent. of those placed in the old lines were already dead of the disease, and that, too, in spite of the fact that the new lines were but 300 yards from the old ones.

Dodds Price next took the further step of moving all the healthy people out of infected houses to new ones on a fresh site. A large number of fresh coolies were also drafted into these new lines. Eighteen months later no case of kala azar had occurred among them, and the lines remained free from kala-azar in subsequent years. This experiment was repeated with good results. In slightly infected lines only the affected households were segregated and their houses burnt, this measure being also of value, although less effective than the former ones.

Attempts were made by Dodds Price to destroy all forms of insect life in the infected houses and those around them in a cooly line in which kala-azar cases continued to arise each cold weather. The houses were fumigated with burning sulphur, the beds thoroughly washed with strong boiling carbolic lotion, the clothes either boiled in the same solution or destroyed, while the blankets were all burnt as most likely to harbour bugs.

All the above measures have not met with any permanent success. Rogers suggested that disinfecting operations, somewhat similar to those just mentioned, should be carried out in endemic areas in houses where kala-azar patients lived. In this way, he thought, much might be done to reduce the spread of the sporadic form of the disease among Anglo-Indians in Calcutta. It does not, however, appear that the disease is getting less common among these people in Calcutta by the adoption of these segregation measures.

The preventive measures that have been adopted in Assam have, until very recently, met with a partial amount of success. The following notes on the preventive measures adopted in Assam are extracted from McCombie Young's KALA-AZAR IN ASSAM (1924). Before the discovery of antimonial treatment, compulsory evacuation of the infected site was the only available preventive measure. Dodds Price adopted measures of removal and segregation of infected individuals in certain tea gardens with apparently successful results. Similar operations were commenced by the Government of Assam, in Golaghat, in 1912. The infected family and patients were removed to a distance of not less than 300 yards from the infected house, and the infected house was burnt down and with it all the less valuable property of the inmates, compensation being paid for their destruction, and any valuable property disinfected. It was, however, found that removal of the infected family alone was not sufficient to eradicate the infection from a village, for in the year after their removal fresh cases of kala-azar were found among the neighbouring houses. Measures were then taken to move, as "contacts," the families who lived in the immediately adjoining houses. This was more successful, but the process had often to be repeated, year after year, as the disease recurred in houses beyond the excised area. In several instances, and particularly in an old-standing infection, it was found that it would have been better to move the whole village in the first instance rather than to move sections of the village in successive years, for the mortality would have been less and the cost the same. At first these operations seemed to give a promising measure of success, and the opinion was expressed in 1916 that "if no new factors arise to vitiate our calculations, we may look for the extinction of the disease in this area within a year or two." This hope was not realized, for a new factor, the influenza epidemic of the cold weather of 1918-19, changed the whole situation, and there was a recrudescence of the disease in areas in which it had been dormant and it threatened to spread to previously uninfected areas. The Government of Assam enacted certain measures of control under the provisions of the Epidemic Diseases Act. The regulations provided for the notification, on the recommendation of the Sanitary Commissioner, of any village area found to be infected with kala-azar, for the prohibition of migration from that area, and for the compulsory removal of any of its inhabitants from an infected site, and for the destruction of the infected house and property. The infected families, that is to say those in which a case of kala-azar was discovered, were grouped in an "infected camp," "contacts," i.e., their neighbours and any other families which for any reason were under suspicion, were located in another group of houses, forming the "contact camp." The remainder were located in a "healthy camp," which was meant to form the nucleus of a new village. There was no migration from notified areas, and the intercommunication with uninfected villages was greatly limited and chiefly confined to visits between relatives. It appears that removal of a community from an infected site and treatment of those among them who are infected with kala-azar would terminate the outbreak in that particular community, but the method is prohibitively expensive upon a large scale. In spite of the effective

prevention of migration from infected areas and limitation of communication between infected and healthy villages, it was found that a gradual diffusion of the disease took place, and year after year a fresh crop of new infections was discovered in previously uninfected villages to which the same expensive measures of removal and control had to be applied. When the general recrudescence which followed the influenza epidemic had to be faced, experience showed that segregation on the scale which would be necessary to deal with a widespread prevalence would be prohibitively expensive and administratively impossible. It was accordingly found necessary to retire to the second line of defence which the introduction of intravenous tartar-emetic treatment had provided.

Treatment as a method of prevention was originally put forward as an alternative to the methods of prevention by segregation by Knowles, in 1920. The results were found to be encouraging. Early treatment of the first one or two cases seemed to control an outbreak. In some cases it appears to extinguish it entirely, perhaps by preventing the establishment of the conditions of site infection, if the first case acquired his infection elsewhere. In practically all cases it seemed to prevent the outbreak assuming extensive proportions. The indications seemed to be that where only one case came under observation and treatment at an early date, there was a reasonable chance that no more cases would be seen. When several cases are seen for the first time, and if they have remained for some time unrecognized and untreated, and the opportunities for the establishment of site infection have been ample, then no amount of treatment of cases seems be able to extinguish an outbreak, and under those circumstances a perennial crop of cases may be expected. It seems that early compulsory treatment has a distinct preventive action where it is efficiently applied in the early stages of a village infection, but removal to a fresh site is still necessary to terminate an infection when it has become deeply rooted by delay in action.

The treatment campaign against kala-azar in Assam has been of immense value as a prophylactic measure. The present-day campaign against the disease in Assam is well described in Health Bulletin No. 9 (Government of India Central Publication Branch, 1927), containing the *Treatment Campaign against Kala-azar, Assam*, as drawn by Major Murison, Director of Public Health, Assam, and the following extracts are made therefrom :—

"The treatment of the disease in Assam with tartar emetic began in 1919, when only a comparatively small number of cases were treated. In the special kala-azar dispensaries and out-centres, sodium antimony tartrate, manufactured by Messrs. Burroughs Wellcome & Company, London, and put up in 'soloid' form, is used exclusively. It is passed through a severe test for purity before being supplied to the Assam Government. Treatment with sodium antimony tartrate, in 1 per cent. solution, given in divided and increasing doses over a period of three months, is the method employed.

"Although treatment with this drug has been very successful, it has the disadvantage of being long and tedious. Treatment is therefore difficult to enforce, as patients who have been completely incapacitated by the disease

improve so considerably after a few injections that they discontinue treatment altogether or attend very irregularly. This irregularity makes it very difficult to effect complete cures. In spite of the regulations in force under the Epidemic Diseases Act to compel patients to undergo a complete course of treatment, our campaign against the disease is being greatly handicapped by the large number of patients who are stopping treatment.

"To overcome this difficulty communiqués are being regularly issued inviting the co-operation of the people. Much propaganda work is being done by means of lantern demonstrations and illustrated posters and pamphlets on the disease, emphasizing the grave dangers of stopping treatment before a complete cure has been effected. This has had some effect in reducing our 'stopped treatment' cases. It was felt that the above difficulties would be still further overcome if some drug could be introduced which was not only as efficacious as sodium antimony tartrate but took a much shorter time to effect a cure."

In 1922 the attention of the Government of Assam was drawn to the most brilliant results obtained by Major Shortt in the treatment of kala-azar, while working under the auspices of the Indian Research Fund Association at the Pasteur Institute, Shillong, by the use of urea stibamine which was sent to him by the present writer at the request of Colonel Greig, Director of Medical research.

After trial, as an experimental measure, the Government of Assam gave sanction in the middle of 1925 to the treatment of all indoor kala-azar patients, and a certain percentage of outdoor patients attending the indoor hospitals, with urea stibamine (Brahmachari) and "471." "The results achieved," the *Bulletin* states, "have proved most satisfactory and encouraging."

"*Instructions for Treatment of Kala-Azar Cases with Urea stibamine (issued in the Bulletin).*

"(1) *Urea stibamine*, an organic compound of antimony, is a pinkish-coloured powder and is usually supplied by the Government of Assam in 1 grm. ampoules, though smaller amounts in capsules are now being issued. The ampoules should be examined for cracks or breaks, and any doubtful ampoules should be rejected.

"(2) *Equipment Necessary.*—(1) Urea stibamine (in sealed ampoules); (2) distilled water; (3) 50-c.c. measures with rubber cap; (4) spirit lamp; (5) test-tube holder; (6) Collins' syringe, 10 c.c. one; (7) rubber tubing for tourniquet; (8) tincture of iodine; (9) sterilizer; (10) cotton-wool.

"(3) *Distilled water* and, if possible, freshly distilled water only must be used in making up the solution of the drug, or serious accidents may occur. Stills or distilled water will be supplied, also test-tubes marked at 50 c.c.

"(4) *To Make the Solution.*—Take one of the marked test-tubes, cleanse it thoroughly in clean boiled water with a brush. If any acid, alkali, chemical or dirt has been allowed to get into the tube, every trace must be carefully removed and the test-tube finally washed with distilled water. Place about 60 c.c. distilled water in the test-tube. Put on a rubber cap over the mouth

of the test-tube. Now perforate the cap with a hypodermic needle and leave the needle sticking through the rubber cap partly in and partly out, but see that the point of the needle does not reach the level of the water inside. Dry the outside of the test-tube to prevent cracking, and gently heat over the flame of the spirit lamp till the water boils quietly for four minutes. The steam will escape through the needle. Stand test-tube to cool. Urea stibamine *must not* be dissolved in hot water. When cool, carefully remove needle and cap with forceps heated in spirit lamp, taking care not to contaminate the boiled water or the edge of the test-tube with the hand or unsterilized instrument. Pour off excess of water down to 50-c.c. mark. Take 1 grm. ampoule, file the neck and break off point. Sterilize the cut end of the ampoule only by passing it through the flame and allow to cool. Pour the whole contents of the ampoule into the test-tube containing the 50 c.c. of cool but recently boiled distilled water, replace the rubber cap aseptically and shake gently till the drug is completely dissolved. This will make a 2 per cent. solution of urea stibamine. The solution must always be freshly made just before use or accidents may occur. The solution should not be heated, and any of the solution not used at the time must be thrown away. If it is desired to make smaller quantities of solution, the amount of water for this may be drawn from the test-tube after it cools into a sterile 10- or 20-c.c. syringe. The rest of the boiled water in the test-tube should be thrown away and the measured quantity in the syringe returned to the marked test-tube. The correct amount of urea stibamine should now be added to make a 2 per cent. solution. The test-tube should always be cleaned with distilled water after use.

"*Dosage.*

"(5) (*a*) For an adult begin with 0·05 grm. and increase by 0·05 grm. until 0·2 or 0·25 grm. at each dose is reached, thus :—

First dose, 0·05 grm. or 2·5 c.c. of the 2 per cent. solution.
Second dose, 0·1 grm. or 5 c.c. of the 2 per cent. solution.
Third dose, 0·15 grm. or 7·5 c.c. of the 2 per cent. solution.
Fourth and every subsequent dose, 0·2 grm. or 10 c.c. of the 2 per cent. solution.

"(*b*) For children between the ages of 5 and 12 years, half the adult dose, thus :—

First dose, 0·025 grm. or 1·25 c.c. of the 2 per cent. solution.
Second dose, 0·05 grm. or 2·5 c.c. of the 2 per cent. solution.
Third dose, 0·075 grm. or 3·75 c.c. of the 2 per cent. solution.
Fourth and every subsequent dose, 0·1 grm. or 5 c.c. of the 2 per cent. solution.

"(*c*) For children between 1 and 5 years of age, one-third the adult dose, thus :—

First dose, 0·01 grm. or ½ c.c. of the 2 per cent. solution.
Second dose, 0·02 grm. or 1 c.c. of the 2 per cent. solution.
Third dose, 0·03 grm. or 1·5 c.c. of the 2 per cent. solution.
Fourth and every subsequent dose, 0·04 grm. or 2 c.c. of the 2 per cent. solution.

"(6) *Frequency of Injection.*—Injections should be given at an interval of one day, e.g., on the first, third, fifth, seventh, &c., or at an interval of two days, whichever may be convenient, e.g., on the first, fourth, seventh, tenth,

thirteenth, sixteenth, &c., day. Treatment must be given at least twice a week regularly to be of value, the first method being preferable.

"(7) *Contra-indications.*—In (1) severe dysentery developing during treatment; (2) marked nephritis with œdema. In both these the dose should be largely reduced. In (3) very advanced cases, begin with a smaller dose, or give at longer intervals. Such special cases are, however, rare.

"*Sterilization of Syringe and Needles.*

"(8) Sterilization is effected at a temperature of 160° C. in melted vaseline.

"*Method.*—Erect folding stand. Place spirit lamp below the opening, and the aluminium dish, three-quarters full of vaseline, on the stand, fix the thermometer in the holder with the bulb dipping into the vaseline. Place the needles to be used in the vaseline.

"When the temperature reaches 100° C., draw up the melted vaseline into the syringe without the needle. Retain for a few seconds and then eject the vaseline back into the sterilizer. When the temperature of the vaseline reaches 160° C. fill and empty the syringe again in the same manner. Pick up the needle from the vaseline with the forceps and fit it on the syringe. Again fill the syringe with the hot vaseline. Eject the vaseline into the sterilizer and turn the syringe needle due upwards. Push down the plunger quickly so as to clear the needle of vaseline. Allow to cool. To ensure that the needle is not blocked with vaseline in cooling, press down the plunger. If the needle is blocked, dip it into the vaseline for a second or two, withdraw it, and then press down the plunger to remove the hot melted vaseline. When the temperature of the vaseline in the sterilizer reaches 160° C. turn down the spirit lamp flame so as to maintain it at this temperature. Record syringes cannot be so sterilized without risk of breaking.

"(9) *Injection.*—Sterilize the top of the india-rubber cap of the test-tube containing the solution by dipping in boiling water. Tilt the test-tube, plunge the needle of the sterilized syringe through the cap and draw into the syringe the required quantity of the solution. Avoid all air bubbles in the syringe. Choose a suitable vein in the patient's arm, sterilize the skin over it with tincture iodine, then proceed to give the injection in an exactly similar manner as with sodium antimony tartrate injections. The patient must be made to remain sitting or lying near the place of injection for at least a quarter of an hour after being injected.

"Concluding Remarks.

"It is a well-known fact that before any treatment for kala-azar was known, at least 90 per cent. of those attacked succumbed to the disease. Taking the figures for the past six years, we find that 185,054 patients suffering from the disease have been treated, of which 166,545 would undoubtedly have died without proper treatment. The benefit conferred on Assam by this

campaign against the disease can be estimated when it is realized that the treatment has converted a death-rate of 90 per cent. into a recovery-rate of the same percentage. We have good reason to hope that we shall one day be successful in stamping the disease out of Assam."

In one of his recent reports on the treatment campaign against kala-azar in Assam, Major Murison points out that the most up-to-date treatment with urea stibamine was now available free of cost to all kala-azar patients and not far from patients' homes. As regards propaganda work, he makes the following remarks: "It is the earnest request of the Public Health Board to the general public to endeavour to make the treatment campaign against the disease more effective by inducing all patients to undergo a complete course of treatment as soon as the disease is detected. An untreated, or a partially treated, kala-azar patient continues to be infectious to others. In the interest of the public health, and in the hope of eradicating the disease, it is therefore the duty of all educated people to induce such patients to undergo a complete course of treatment as early as possible."

No speculative suggestions are made by me in this treatise for the destruction of, or protection against, sandflies as a prophylactic measure, since they may be as valueless as any that could have been formulated a few years ago for the destruction of bed-bugs, which were once supposed to be the carriers of the disease. Instead of asking the poor patient to adopt measures which may be expensive for him to carry out, and subsequently turn out to be ineffective, we would like to wait till the transmission problem is solved.

(B) Mediterranean Countries.

No organized prophylactic measures have hitherto been instituted. Lignos advocated the destruction of all the dogs on the island of Hydra. The researches of Massaglia, Marshall, Pereira da Silva, Gabbi, and many others have shown that the flea can play no part in the transmission of the disease. Destruction of dogs in an infected area may, therefore, be of doubtful value. Basile, however, claimed that in Bordonaro, in Sicily, where a high percentage of naturally infected dogs occurred, the extermination of these animals led to an almost complete disappearance of the human disease (1916).

The apparently beneficial results following measures for the destruction of bugs may be due to the destruction of some other unrecognized blood-sucking insect which may be the true carrier of the disease. If this turns out to be the sandfly, then measures against it will have to be devised.

The fact that a previous attack of the disease confers an immunity in a large percentage of cases, and that the disease spreads slowly from one place to another even when showing epidemic manifestations, makes it possible that an intensive mass treatment with a specific should lead to its eradication. This is the basis of the present-day campaign against kala-azar in Assam.

For a specific to be useful in prophylaxis, it should primarily be of very high therapeutic value. It must also be very quick in its action, so as to reduce

the chances of infection from one person to another, and to avoid the disadvantages of long and tedious treatments. Such a specific for kala-azar has been found in urea stibamine, as extensive observations in campaigns against kala-azar have conclusively proved.

The following extract from the speech of His Excellency Sir John Kerr, while bidding farewell to the second Legislative Council in Assam (1926), shows the value of this campaign. After referring to the value of the treatment of kala-azar with urea stibamine, His Excellency said : "We may now say that victory, if not in sight, is assured. The progress in the campaign against kala-azar in Assam has been phenomenally rapid, and if it continues at the present rate there is an excellent prospect of the dread scourge being brought under complete control in a few years."

APPENDIXES.

I.

LABORATORY METHODS.

No attempt will be made to discuss in detail the various laboratory methods and apparatus here referred to. Only a short account will be given of them with special reference to what are necessary in the diagnosis of kala-azar. For details the reader should consult books on clinical laboratory diagnostic methods and on hæmatology.

ESTIMATION OF HÆMOGLOBIN OF THE BLOOD.

The most exact method is by the estimation of the amount of iron (dry hæmoglobin containing 0·42 per cent. of iron) in the ash of a given specimen of blood, but this is a somewhat complicated process and is hardly applicable in general practice or clinical work.

For bedside estimation *Tallquist's scale* is frequently used. It consists of a scale of colours and blotting paper strips to suck up blood for examination. The finger or the ear-lobe is sterilized by means of absolute alcohol, which is then allowed to dry. It is pricked by means of a lancet shaped sharp needle with a firm quick stab and, if possible, no pressure should be exerted. The first drop of blood is wiped away. The blood is then sucked up in one of the strips of blotting paper and allowed to dry on it, and the tint obtained is compared with the scale by direct light and the hæmoglobin thereby estimated.

For more accurate estimation Haldane's modification of Gower's hæmoglobinometer may be used, if coal gas is available. The standard of comparison is a hermetically sealed tube filled with a solution of hæmoglobin saturated with carbon monoxide gas. This keeps unchanged for years. A few drops of distilled water are placed in the graduated measuring tube supplied with the instrument. A small quantity of blood is drawn up to the graduated mark in the pipette supplied for the purpose, and is then ejected into and mixed with the distilled water in the measuring tube, and a stream of coal gas is then passed through the blood by means of a cap for attachment to the gas-burner. This converts the hæmoglobin present in the blood into carbon monoxide hæmoglobin. It is then diluted with water to match the standard.

As coal gas is not always available, the instrument is frequently supplied with an additional standard tube containing picro-carmine jelly, as in the original Gower's hæmoglobinometer. This is liable to slow alteration in keeping. It should be kept in the box and not exposed unnecessarily to light, to avoid alteration.

In Sahli's instrument, a standard solution of acid hæmatin is used for the purpose of comparison. This is claimed by some observers to keep even better than the carbon monoxide hæmoglobin solution used in Haldane's apparatus, but others deny this.

Among other hæmoglobinometers may be mentioned the original Gower's, Fleischl's and its modification by Meischer, Kuttner's modification of Sahli's, Oliver's, Dare's, Newcomer's, &c.

ENUMERATION OF BLOOD-CORPUSCLES.

This is effected by means of a hæmacytometer. It consists of a counting slide with a chamber and two pipettes for diluting the blood for the purpose of counting the erythrocytes and leucocytes, which may be conveniently named erythrocytometer and leucocytometer respectively. Slides with various kinds of counting chambers have been devised, of which two are mentioned here, the Thoma-Zeiss or the closed chamber, and Bürker, or open with two counting chambers side by side for counting erythrocytes and leucocytes. The first one is used by most workers in India, while the latter is very popular in America. Similarly, various modifications of the rulings of the slides have been devised. The Neubauer ruling with the Thoma ruling in the centre is very convenient.

The following formula is applied in estimating the number of corpuscles in one c.mm. of blood with Thoma-Zeiss hæmacytometer or with one having similar ruling :—

No. of corpuscles $= \frac{4,000 \times A \times Z}{N}$, where A is the number of corpuscles counted, Z is the dilution used, and N is the number of squares counted.

From the above formula a simple method for counting the corpuscles can be deduced, which consists of adding four ciphers to the number of cells present in eighty small squares in a dilution of 1 : 200, or in forty small squares in a dilution of 1 : 100.

Generally speaking, it may be laid down that the higher the dilution and at the same time the greater the number of squares counted, the more accurate is the counting.

The dilution of the blood frequently employed for counting the erythrocytes is 1 : 200, but in cases of severe anæmia it is more convenient to use a dilution of 1 : 100.

Ordinarily, the leucocytes may be counted with either of the two pipettes described above. The erythrocytometer is sufficiently accurate in cases in which there is no leucopenia or the leucocyte count is high, as in cases of leukæmia or other diseases in which there is leucocytosis. In the case of leukæmia it is disadvantageous to use the leucocytometer.

On the other hand, in cases of kala-azar and certain other conditions, the number of leucocytes present in the blood may be so low that the latter pipette is absolutely necessary for counting them. Many people use only the ruled area in counting the leucocytes (256 squares in Thoma-Zeiss), the process being repeated upon a second and third slide, and the average taken. With Neubauer and similar rulings and a dilution of 1 : 20, a convenient plan is to

PLATE XII.

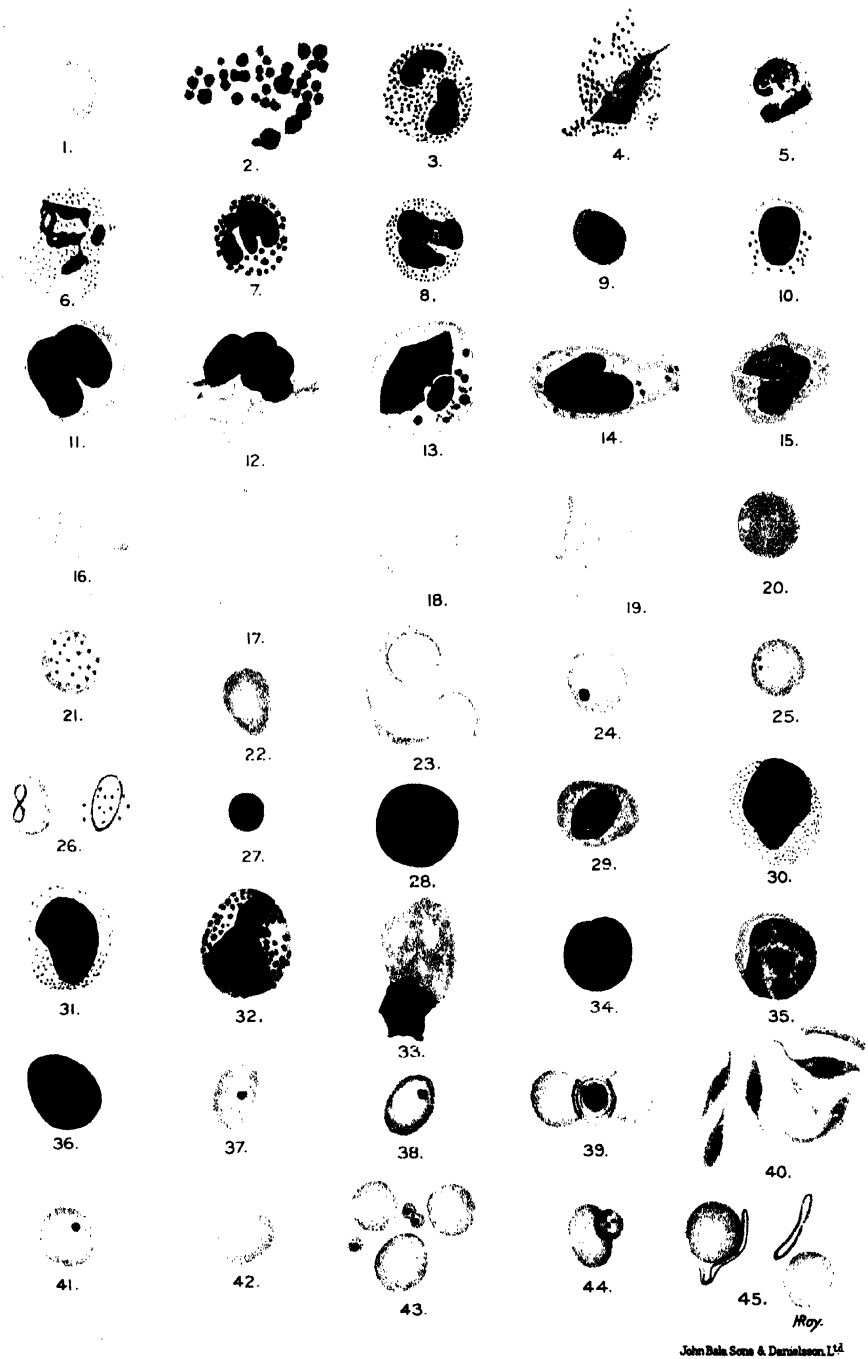
Normal and Abnormal Blood-Cells, &c.

(Partly original and partly from Schleip's "Hæmatological Atlas," 1920; Panton's "Clinical Pathology," 1913; and Byam and Archibald's "Practice of Medicine in the Tropics," 1921.)

Fig. 1. Normal red blood-corpuscle.
„ 2. Blood platelets.
„ 3. Coarsely granular eosinophile leucocyte.
„ 4. The same, ruptured.
„ 5. Polymorphonuclear leucocyte.
„ 6. The same, ruptured.
„ 7. Coarsely granular basophile leucocyte, or mast cell.
„ 8. Finely granular basophile leucocyte.
„ 9. Small lymphocyte.
„ 10. Large lymphocyte.
„ 11. Large hyaline mononuclear leucocyte.
„ 12. The same, ruptured.
„ 13. Large hyaline mononuclear leucocyte, containing malarial hæmozoin pigment.
„ 14. The same.
„ 15. Transitional mononuclear leucocyte.
„ 16. Poikilocytosis.
„ 17. Microcytes.
„ 18. Macrocyte.
„ 19. Achromia.
„ 20. Polychromatophilia.
„ 21. Punctate basophilia.
„ 22. Diffuse basophilia.
Fig. 23. Demilune red blood-corpuscles.
„ 24. Howell-Jolly body.
„ 25. Chromatin dust in red blood-corpuscle.
„ 26. Cabot's rings.
„ 27. Normoblast.
„ 28. Megaloblast.
„ 29. Myeloblast.
„ 30. Neutrophile myelocyte.
„ 31. Eosinophile myelocyte.
„ 32. Basophile myelocyte.
„ 33. Endothelial cell.
„ 34. Türck cell.
„ 35. Lymphoblast.
„ 36. Rieder's cell.
Figs. 37-38. Platelets lying on red corpuscles.
Fig. 39. X-bodies of Horrocks and Howell (1908).
„ 40. Distorted platelets.
„ 41. Deposit of stain on a red corpuscle.
„ 42. Vacuolated red corpuscle.
„ 43. Yeasts on the slide.
„ 44. A protozoon on the slide from an extraneous source.
„ 45. Pessary bodies, from destroyed red corpuscles.

(*From* Knowles' and Senior-White's "Malaria: its Investigation and Control." *By kind permission of Major Knowles.*)

PLATE XII.



HRoy.

John Bale, Sons & Danielsson, Ltd.

count the leucocytes with a low power objective in the square millimetres at each of the four corners of the ruled area, and multiply by 50.

While it is more convenient to estimate the number of leucocytes over the squares in conditions in which the leucocytes are numerous, it is very desirable in cases of kala-azar and similar conditions with low leucocyte count to count the leucocytes in the field of vision instead of in the ruled areas, care being taken to go regularly from one circular area to another, and to include an entirely fresh field at each count. The areas counted must not be selected, but should be taken just as they come.

The number of squares in the field of vision is calculated from its area, which is easily determined from the formula : Area = πr^2 where r is half the diameter of the field of vision. The sliding tube of the microscope is drawn to a point at which the diameter of the field of vision is just sufficient to equal eight or ten times the length of the side of a small square on the counting slide.

To ensure that a diameter is being measured is easily determined by observing that it is the greatest of all the chords of the field of vision parallel to it, or by noticing that a chord drawn at right angles to it from its middle point contains the same number of sides of the small squares as the diameter itself, and therefore is also a diameter.

The number of leucocytes present in 1 c.mm. is then calculated from the formula already given for estimating the number of corpuscles.

Various fluids have been used for diluting the blood for examination with the hæmacytometer. A few of these are mentioned here :—

Gower's Solution.—Sodii sulph., 104 gr. ; acetic acid, 1 dr. ; distilled water 6 oz.

Hayem's Solution.—Sodii chloride, 1 grm. ; sodii sulph., 5 grm. ; corrosive sublimate 0.5 grm. ; distilled water, 200 c.c.

Toisson's Fluid.—Methyl violet, 0·025 grm. ; sodii chloride, 1 grm. ; sodii sulph., 8 grm. ; neut. glycerine, 30 grm. ; aqua distill., 160 grm.

The diluting fluid for counting leucocytes by means of the leucocytometer should dissolve the red corpuscles so that they will not obscure the leucocytes. The simplest fluid is a 1 per cent. solution of acetic acid. More satisfactory is the following : Glacial acetic acid 1 c.c. ; 1 per cent. aqueous solution of gentian-violet, 1 c.c. ; distilled water 100 c.c. (These solutions must be filtered frequently to remove yeasts and moulds.)

Among other types of counting chambers and rulings of the slides may be mentioned the original Gower's counting chamber, Levy counting chamber, Hausser's counting chamber, and Zappert's rulings of the slides.

For cytological work other than blood-counting, as in cerebro-spinal fluids, &c., the Fuchs-Rosenthal ruling may be used. For the counting of blood-platelets, the so-called Helber chamber with a Neubauer ruling, as for blood-counting, may be used.

REFERENCES.

ROGER S. MORRIS, M.D. "The Choice of Ruling in a Blood-Counting Chamber," *Journal of Laboratory and Clinical Medicine*, December, 1916, vol. ii, p. 208.

HORACE GRAY, M.D. "Cell Counting Technic : A Study of Priority," *American Journal of the Medical Sciences*, October, 1921, No. 4, vol. clxii, p. 526.

STUDY OF STAINED BLOOD.

For making proper films, it is essential that the slides should be perfectly clean. When new slides are used, the superficial grease is removed by breathing on them or putting a little alcohol and rubbing up with a clean handkerchief, or by flaming them. Used slides and cover-glasses are placed first in a mixture of concentrated sulphuric acid 6 parts, potassium bichromate 6 parts, and water 100 parts, for some time. They are then thoroughly washed and kept in absolute alcohol after drying. When required for use they are to be taken from the alcohol, and when all the alcohol has drained off they should be rubbed with a clean handkerchief or passed through a flame.

After the preliminary preparation for taking blood which has been described before, a droplet of blood is taken on a clean slide a little away from one of its ends, taking care that this slide does not touch the skin. The edge of a second slide is placed against the surface of the first at an angle of about 45° touching the drop of bood, and the blood allowed to spread itself at the junction of the two slides. The second slide is now pushed at this angle to the opposite end of the first, when an evenly spread film is formed. The blood should follow the spreading slide, and not be pushed in front of it. The thickness of the film depends on the angle between the two slides, the size of the drop of blood and the pressure between the slides. The film is now dried in the air or by holding it high above a flame. It is then stained by one of the modifications of Romanowsky stains, such as Leishman's or Wright's modifications of it, or Giemsa's, and then examined under the microscope with oil-immersion. The author generally uses Wright's stain, and he finds that it gives most satisfactory results.

In stained specimens of the peripheral blood, apart from examining for the presence of Leishman-Donovan bodies, the following diagnostic information indicative of disease may be obtained from this examination :—

(1) *Erythrocytes.*—Pathologically, they vary in hæmoglobin content, size and shape, staining properties and structure. The depth of staining furnishes a rough guide to the hæmoglobin content of the corpuscles. Variations in size (anisocytosis) are shown by the presence of microcytes, macrocytes and megalocytes. Variations in shape are shown by the presence of poikilocytes, such as oval, pyriform and club-shaped cells, &c. (poikilocytosis). Variation in staining properties is shown by polychromatophilia, basophilic granular degeneration, &c. Variation in structure consists in the presence of nucleated red cells or erythroblasts. They include the microblasts, normoblasts and megaloblasts.

(2) *Leucocytes.*—The differential count of leucocytes yields more helpful information than any other single procedure in blood examination. It is best made on a film stained with one of the Romanowsky stains. The abnormal varieties of leucocytes include (*a*) myelocytes ; (*b*) myeloblasts ; (*c*) lymphoblasts ; (*d*) Türck's irritation leucocytes ; (*e*) plasma cells ; (*f*) degenerated forms ; and (*g*) atypical forms.

To make the differential count, it is necessary to go carefully over the

film with an oil-immersion lens, using a mechanical stage, if available. After classifying each leucocyte seen, the percentage of each variety, in the total number of leucocytes counted, is calculated. For accuracy, 500 to 1,000 leucocytes should be classified; for approximate results, 300 are sufficient, but it is imperative to count the leucocytes in all parts of the smear, since the different varieties may not be evenly distributed.

The actual number of each variety in 1 c.c. of blood is easily calculated from these percentages and the total leucocyte count. An increase in actual number is an *absolute increase*; an increase in percentage only, a *relative increase*. It is evident that an absolute increase of any variety may be accompanied by a relative decrease.

To get a fairly uniform distribution of leucocytes in a film made on a slide for differential count of leucocytes, it must not be made very thin, and should be moderately thick, but not so thick as to make the leucocytes difficult to be recognized.

Suggestions have been made from time to time to carry out the differential count in the counting chamber along with the total count by use of diluting fluids which colour the leucocytes differentially. As special diluting fluid for this purpose Stitt uses 1·5 c.c. of neutralized formalin in 98·5 c.c. of 0·5 per cent. glycerol. Just before use this fluid is coloured with Giemsa's stain by adding one drop of the stain for each 1 c.c. of the fluid. Formalin may be neutralized by adding a few drops of phenolphthalein, and then adding very dilute sodium hydroxide to the first appearance of a pink colour.

METHOD OF FINDING LEISHMAN-DONOVAN BODIES IN THE PERIPHERAL BLOOD.

(1) *Thin Film Method.*—In a thin film of blood prepared in the way indicated above, in order to search for the Leishman-Donovan bodies, the edges of the film and the tail where the leucocytes tend to specially gather should be examined. The parasites are found either inside the polymorphonuclears or the mononuclears, or very rarely apparently free, due to rupture of the cells containing them in the process of making the smear. By compressing the finger below the pulp for half a minute or so and then puncturing it, a larger number of leucocytes may be obtained in the smear.

In kala-azar leucopenia is frequently present, and oftentimes to a marked extent, and it is therefore very difficult to find, in thin films of the peripheral blood, a sufficiently large number of leucocytes for examination for the detection of Leishman-Donovan bodies inside them. It has therefore been recommended to examine thick films for this purpose.

(2) *Thick Film Method.*—Ross's thick film is made by allowing some six drops of blood to fall on a slide within an area 5 to 7 mm. in diameter and spreading them into an even layer. After dehæmoglobinization with water the resulting film is dried in air and stained.

Reference has already been made to the thick film method of Knowles and Das Gupta (p. 84). Care should be taken to let the films dry in the air as completely as possible, so that the leucocytes may adhere to the slide and not be washed away during subsequent manipulation.

Another method is to take one-half to one drop of blood, which is made into a thick film by spreading it out somewhat with the corner of another slide. Usually half a drop of blood will make a spread of proper thickness about half an inch in diameter. It is then dehæmoglobinized and fixed in methyl alcohol (Merck's highest purity), containing 2 per cent. glacial acetic acid and 2 per cent. hydrochloric acid. It takes from one to three minutes to decolorize recently-made preparations. Wash, preferably in running water, to entirely remove the acid. This requires only a minute or two, and then stain. (For other methods see Addendum.)

(3) *Centrifuge Method.*—The author has adopted the following method for concentrating the leucocytes from the peripheral blood: About 50 c.mm. of blood are centrifugalized either alone or after mixing with a 2 per cent. solution of sodium citrate to prevent clotting, and after pipetting off the supernatant layer of fluid, the upper leucocyte layer and the upper quarter or so of the column of red corpuscles are pipetted off and spread on a slide and stained. In this way there is a great preponderance of leucocytes in every field of the microscope. The author has sometimes concentrated the leucocytes by putting blood mixed with a little sodium citrate solution into miniature tubes closed at one end and then centrifuging in a hæmatocrite. The leucocyte layer at the top of the sediment at the bottom of the tube is examined in the way described above. By centrifuging 5 c.c. of blood mixed with Locke's solution (sodium chloride 9·2 grm., potassium chloride 0·5 grm., calcium chloride 0·1 grm., sodium citrate 10 grm., distilled water 1,000 c.c.), at a speed of 750 revolutions, Young and Van Sant state that parasites are readily discovered in films made from the bottom-most part of the centrifuging tube inside mononuclear and polymorphonuclear leucocytes. They hold that these leucocytes are heavier than other blood-cells when they contain Leishman-Donovan bodies, and therefore go to the bottom of the centrifuging tube during centrifugalization. If the cloudy layer at the top is decanted and centrifuged at a high speed, the extra-corpuscular Leishman-Donovan bodies may be found in the blood-platelet layer of the sediment.

In the author's experience the search for the parasite in the peripheral blood takes an extremely long time and is frequently negative, whatever may be the process adopted, and in one case he examined at least 400 leucocytes without finding any Leishman-Donovan bodies in them.

METHOD OF STAINING BLOOD FILMS.

This is effected by one of the Romanowsky stains, which consist of a compound containing a mixture of methylene-blue and its derivatives in combination with eosin, there occurring between the methylene-blue and the eosin some complicated chemical reactions. Modifications of Romanowsky stains chiefly used are :—

(1) Giemsa's stain.
(2) Leishman's stain.
(3) Wright's modification of Leishman's stain.
(4) Jenner's stain.

These stains can be obtained from Grubler or Merck, Germany, or from English firms, such as Burroughs Wellcome and Co., or British Drug Houses, Ltd.

For the preparation of Giemsa, Azur II eosin, 3 grm., and Azur II, 0·8 grm., are put in a mortar and then dissolved in glycerine (chemically pure, Merck), 250 c.c., by means of a pestle. Then add 250 c.c. of methyl alcohol (Merck's reagent).

Leishman's stain may be prepared easily by dissolving 0·15 grm. of Leishman's powder in 100 c.c. of acetone-free methyl alcohol.

Wright's stain is prepared by dissolving 0·1 grm. of the powder in 60 c.c. of pure acetone-free methyl alcohol. His most recent directions for its preparation are as follows :—

To a 0·5 per cent. aqueous solution of sodium bicarbonate add (B. X or medicinally pure) methylene-blue in the proportion of 1 grm. of the dye to each 100 c.c. of the solution. Heat the mixture in a flask in a steam sterilizer at 100°C. for one full hour. After heating, allow the mixture to cool, and then filter it to remove the precipitate which has formed in it. To each 100 c.c. of the filtered mixture add 500 c.c. of a 0·1 per cent. aqueous solution of "yellowish water-soluble" eosin, and mix thoroughly. Collect the abundant precipitate, which immediately appears, on a filter. Dry the precipitate thoroughly, dissolve it in methyl alcohol (Merck's "reagent") in the proportion of 0·1 grm. to 60 c.c. of the alcohol. In order to facilitate solution, the precipitate is to be rubbed up with the alcohol in a porcelain dish or mortar with a spatula or pestle. This alcoholic solution of the precipitate is the staining fluid.

In staining with Giemsa's stain, the film is fixed in pure methyl alcohol; then Giemsa's stain, diluted with distilled water in such a way that one drop of stain is mixed with 1 c.c. of water, is poured over it and allowed to act for fifteen or twenty minutes.

In staining with Leishman's stain, no preliminary fixing of the film is required, it being effected by the methyl alcohol contained in the solution. A few drops of the undiluted stain is poured and kept over the film for about half to one minute, taking care that it does not dry. The solution is then diluted by dropping about double the quantity of fresh distilled water and the stain and the water are quickly mixed with a glass rod. Five minutes are now allowed for staining, and the stain is then gently washed off with distilled water. A little of the water is kept on the film for half a minute to intensify the colour contrasts in the various cells.

In staining with Wright's stain, cover the film, without previous fixation, with plenty of the staining fluid by means of a dropper, so as to avoid too great evaporation and consequent precipitation. After one minute add to the staining fluid on the film the same quantity of distilled water by means of a second dropper. Allow the mixture to remain for three to six minutes, according to the intensity of the staining desired. Wash the slide in water for thirty seconds or until the thinner portions of the film become yellow or pink in colour. Dry with filter paper or by waving high above a flame.

PERIPHERAL BLOOD-CULTURE.

In a series of 440 undoubted cases of kala-azar observed by Brahmachrai and Maity, the peripheral blood-culture was found by them to be positive in 426, i.e., 97 per cent. In another series of 190 cases in which cultures were made from material obtained on spleen puncture, it was found positive in all. The technique of blood-culture has been described (p. 86). A few further points are mentioned here.

A 2 c.c. all-glass sterilized syringe is used for puncturing a vein in the arm. Sterilization of the syringe with the needle is effected by autoclaving the syringe in steam, and subsequently removing all the traces of water inside the barrel and the needle by one up-and-down movement of the piston through the barrel, the needle being attached to the syringe by means of a pair of forceps. They may also be thoroughly dried in a hot-air sterilizer. The method of the Kala-Azar Commission is to use a 5 c.c. all-glass syringe, and to put up the syringe, in their metal cases wrapped in paper and autoclaving. They are afterwards thoroughly dried, without opening the cover, in a hot-air sterilizer. Oil sterilization is hardly necessary. The oil is likely to remain sticking inside the syringe and thereby collect dirt after repeated use, which is frequently difficult to clean, especially in an all-metal syringe, the inside of which cannot be seen.

The technique of venipuncture has already been described under the chapter on metallic antimony (p. 116). The best method of making the vein prominent is by means of an india-rubber tubing tightened round the arm above the point of puncture by means of a pair of tongue forceps, in the manner indicated in the diagram. By opening the forceps and tightening or loosening the rubber tubing, the amount of compression of the arm may be very easily regulated so as to stop the venous flow to such an extent as may be desired. The compression should not be so great as to stop the arterial supply. Subsequently the blood drawn is dealt with in the manner described in p. 86. The blood should be diluted with sterile sodium citrate solution. It is desirable that the blood should be properly diluted in citrated saline for successful culture. This washes out substances in the blood-plasma that may be inimical to growth. The Kala-azar Commission point out that this dilution with citrate solution is important, not only as assisting manipulation, but as favouring culture by the absence of clot formations in the medium and possibly through the anticomplementary action of the salt. It is desirable that only freshly prepared medium (up to seven days after preparation) should be used and culture kept at 22° C. to 24° C.

Row describes the following technique for what he calls "intensive culture" from the peripheral blood. A few drops of blood from the finger are introduced directly into 15 to 20 c.c. of citrated saline before any clotting takes place. The diluted serum is then centrifuged and the sediment of corpuscles planted directly into a suitable medium, e.g., N N N medium. In this way a growth of flagellates may be obtained in six days. (See p. 86.)

In some cases the author has obtained positive flagellate culture as early as the third day, but generally it takes place in seven to ten days. One should

not consider that the culture is negative until three weeks have passed after incubation, and no growth has been obtained in this period.

The flagellates should be looked for in the fluid of condensation and in the upper layer of the solid portion of the N N N medium in contact with the fluid. The fluid and a portion of the solid should be pipetted off with a sterile capillary tube for examination, and the culture tube put back into the incubator for flagellate growth, if it has not already taken place. A drop of the pipetted material is put over a slide and a cover-glass placed over it, and the wet specimen examined with objective 40 and eye-piece 10 (new notation); with an oil-immersion (90 or 100 new notation), a lower eye-piece 5 (new notation) should be used.

Christophers, Shortt and Barraud have employed various devices for studying the growth of the flagellates in culture. The following are quoted from their observations: "In order to study the life cycle we have employed various devices, such as growth on solid medium (plates) and various forms of microculture. Particularly interesting results have been obtained by taking cover-glass impressions (wet and dry fixed) from the surface of N N N medium plates. These plates are inseminated by placing a drop or two of the citrated infective material in the syringe upon the surface of the medium, covering and tilting the plate so that the fluid flows for a short distance over the medium. The entire track of the fluid can then later be removed as an impression on a 2 in. by $\frac{3}{4}$ in. cover-glass, giving a preparation in which the development of the parasite can be studied *in situ*.

"In addition to observations made in hanging-drop preparations, we have obtained useful results by a form of microculture put up in the following way: A series of sterile slides is prepared with vaseline rings. These rings must be of uniform thickness, and are best made with the aid of a thin glass tube with smooth edge of the required diameter dipped into melted paraffin. On the slide, in the centre of the vaseline ring, is placed with sterile precautions a drop of culture medium (condensation fluid of N N N medium) To each drop of culture medium is then added a drop of material containing flagellates, or the drops on the series of slides are used to make a dilution series, the inseminating material being added only to the first drop of the series. Each preparation is now covered with a sterile cover-glass, which is pressed down to meet the fluid, care being taken to keep the cover-glass strictly parallel with the slide. The result of this procedure is a disc of fluid lying between the cover-glass and the slide, surrounded by air, in the centre of a sealed vaseline ring. The thickness of the disc can be regulated by pressure; it should be of such a thickness as to admit of its being examined throughout its depth by a sixth objective. If the vaseline seal be made of uniform thickness and the cover-glass evenly pressed down, the disc of fluid will remain unchanged in shape or position for weeks.

"Preparations can also be made to include a thin rectangular slab of the solid portion of the medium. This is done by scoring with a sterile needle a specially prepared very thin N N N plate, or selected portion of such, with two series of lines at right angles, the lines of each series being about 1·0 mm.

or less apart. One or more of the small rectangles of medium so formed are removed with a platinum loop to the drop from which the microculture is to be made. The inclusion of the solid medium enables the behaviour of the parasite in relation to the solid parts of the culture medium to be studied, and also facilitates the mapping of the preparation."

STAINING OF LEISHMANIA.

For staining the *Leishmania*, the author generally uses Wright's modification of Leishman's stain. Shortt's method of staining the flagellates has already been described (p. 26) and gives very good results.

For dried preparations the ordinary method of fixation in absolute alcohol may be employed.

When making preparations for Giemsa's stain from N N N medium, it is necessary that the culture fluid should be diluted with five or more parts of citrate solution. Washing by centrifuging gives very good results for ensuring proper staining of the flagellum with Giemsa's stain.

Christophers, Shortt and Barraud recommend that the film, after having been *dried*, should be exposed for a short period (five to ten seconds) to the vapour of osmic acid.

For wet fixed films, Christophers, Shortt and Barraud have compared the results obtained by fixation in: (1) acetic-sublimate; (2) alcohol-sublimate (Schaudinn); (3) alcohol-sublimate in saline (Maier); (4) picro-acetic-formol (Bouin); (5) alcohol-picro-acetic-formol (Duboscq-Brasil); (6) picro-acetic-formol with neutral copper acetate to increase the solubility of the picric acid (Bouin-Hollande); and (7) osmic-acetic-chromic (Fleming). Of these, they got the best results with Nos. 2, 3, 4 and 6, and after various trials they used No. 2 as a routine practice.

STUDY OF STAINED FILM FROM MATERIAL OBTAINED FROM TISSUES IN KALA-AZAR.

The Spleen.—We have already described the technique of spleen puncture. Its dangers though only slight still exist. The operation has not been popular in private practice in endemic areas, and even in Calcutta, and the author cannot recommend that it should be promiscuously undertaken in all cases of kala-azar, especially in out-patient departments of a large hospital, where it may be impossible to determine all the contra-indications in a short time. An accident following such an apparently simple operation may be unexpected and make the treatment very unpopular. Though the author has not met with any fatal cases in several hundreds of cases in which he or his assistants performed it, yet he is aware that it did turn out fatal in the hands of very competent men. In one or two of his cases in private practice, the patient complained of pain over the abdomen, which became very severe a few hours after puncture and continued for a few days, and there is no doubt that there was hæmorrhage from the seat of puncture which fortunately did not turn out fatal.

In a successful puncture the contents of the needle should practically

consist of spleen pulp only. If the barrel of the syringe shows the presence of blood, it indicates that there has been an undue dilution of splenic material with blood, which is disadvantageous to culture as well as to finding Leishman-Donovan bodies in smears made therefrom, and it is not infrequently observed that a minute quantity of splenic pulp shows Leishman-Donovan bodies in almost every field, while a smear made from the contents of the syringe containing a large quantity of blood from the spleen of the same case may show very few or no Leishman-Donovan bodies. We have stated before that, according to Christophers, in certain stages of the disease the *Leishmania* may disappear from certain portions of the spleen. We agree with this view. In a series of 190 cases of kala-azar, the smear from spleen-puncture material was positive in 185, and the culture from the same was positive in all.

Liver.—It has been recommended that, to avoid the dangers of fatal haemorrhage from spleen puncture, the liver should be punctured by preference, as it is less easily torn than the spleen. But smears from liver-puncture material are more likely to give negative results than from spleen puncture.

FLAGELLATE CULTURE OF SPLENIC OR LIVER MATERIAL.

This is done in the same way as culture from the peripheral blood, a small amount of material being obtained by means of a syringe and diluted with 1 to 2 c.c. of a 2 per cent. sodium citrate solution, and then put into N N N medium. In making a culture, liver puncture should be made in those cases in which there are contra-indications in puncturing the spleen.

Though it may be argued that cases of fatal haemorrhage have been met with after exploratory puncture of the liver in cases of suspected hepatic abscess, yet in all cases in which the author has observed such an accident, very stout and long exploring needles were inserted very deep into the substance of the liver several times for discovering pus, and on post-mortem examination one of the large branches of the portal vein was found to have been punctured. On the other hand, in making punctures for getting a smear for searching for Leishman-Donovan bodies, or for making a culture, a fine needle is required, and there is no danger in puncturing the liver with such a needle.

Smears from other Tissues.—Smears from material obtained from bone puncture, excised lymphatic glands, scrapings from bases of artificially produced blisters or ulcers, have not given satisfactory results in the hands of the author.

BIOCHEMICAL TESTS IN KALA-AZAR.

We have already mentioned the biochemical reactions of the blood in kala-azar, and shall refer here to those points that may be used for diagnostic purposes;—

Relative Haemoglobin Value of Resistant Erythrocytes.—As early as 1909, Brahmachari showed that the *relative haemoglobin value of the resistant erythrocytes* during haemolysis of blood with hyposmatic saline (N : 50) or

distilled water was less in kala-azar than in the case of other diseases or in health.

By *resistant erythrocytes* are meant erythrocytes that do not hæmolyse when one part of blood is treated with two parts of N : 50 saline or distilled water. The *relative hæmoglobin-value* is expressed as the quotient obtained by dividing the amount of hæmoglobin in the *resistant corpuscles* by that of the total blood, and is estimated by the following method : After thoroughly mixing 5 c.mm. of the blood with 10 c.mm. of N : 50 saline, the mixture is centrifuged as thoroughly as possible, and then the sediment is washed several times with N : 10 saline till the supernatant fluid at the top is perfectly colourless. The sediment is now dissolved in a small quantity of distilled water with the addition of a drop or two of chloroform, and then the amount of hæmoglobin is estimated by a Haldane's hæmoglobinometer. In those cases in which the amount of hæmoglobin in the resistant corpuscles is less than 10 per cent., 10 or 20 c.mm. of blood is taken and then treated with 20 or 40 c.mm. of N : 50 saline respectively, and the amount of hæmoglobin in the resistant corpuscles is then estimated. This number, divided by 2 or 4, as the case may be, gives the amount of hæmoglobin in the resistant corpuscles of 5 c.mm. of blood. The amount of hæmoglobin present in 5 c.mm. of blood is then estimated in the ordinary way by means of the hæmoglobinometer.

The author has found the above factor to be, in the average, 0·260 in the case of kala-azar and 0·426 in the case of healthy students. In the case of ankylostomiasis it may be as high as 0·714.

THE GLOBULINS IN KALA-AZAR.

The globulins in kala-azar blood-serum were first investigated by Brahmachari as early as 1917. When human serum is diluted with excess of distilled water it becomes cloudy, owing to partial precipitation of serum globulin. Under certain circumstances a copious precipitate forms instead of a mere cloudiness.

Upon the above observation depend the *globulin ring and the globulin precipitation tests* of the author, which are described below : —

The Globulin Ring Test.—2 c.c. of the blood from a prominent vein of a kala-azar case are drawn by a glass syringe and the blood quickly centrifuged. The serum freed from the clot is introduced into a miniature test-tube with a capillary pipette, and then a small amount of distilled water is gently poured over the serum. A distinct *white ring* forms over the surface of the serum in every case of kala-azar. This is the "*globulin ring test*" of kala-azar. This test is positive even when the blood is diluted ten to twenty times with normal saline.

The Globulin Precipitation Test.—The serum is collected inside miniature test-tubes and then mixed with two or three parts of distilled water. A white precipitate forms in every case of kala-azar. This is the "*globulin precipitation test*" of kala-azar. This precipitation may be due either to the presence of excess of globulin in kala-azar serum, or a specific globulin more easily precipitable than normal serum globulin.

Anti-complementary Globulin Test.—The author has also found that the above globulin has marked anti-complementary properties, and upon this depends the anti-complementary globulin test which is described as follows : 1 c.c. of the serum of a kala-azar case is mixed with 15 c.c. of distilled water. The precipitate is collected and then washed thoroughly with distilled water. The precipitate is then dissolved in 1 c.c. of normal saline and the following observations made : (1) Take, for instance, a hæmolytic system in which the following are the doses of the component parts—0·15 c.c. of anti-sheep amboceptor + 0·5 c.c. of guinea-pig's complement + 0·5 c.c. of sheep's corpuscles = complete hæmolysis. (2) (*a*) Mix 0·5 c.c. of the guinea-pig's complement with 0·2 c.c. of the solution of the above precipitate in normal saline—incubate for half an hour ; (*b*) add to this 0·15 c.c. of anti-sheep amboceptor + 0·5 c.c. of sheep's erythrocytes = no hæmolysis. (3) 1 c.c. of the serum of the same kala-azar case + 0·5 c.c. of sheep's erythrocytes = complete hæmolysis (due, no doubt, to the natural complement and amboceptor frequently present in human serum). (4) (*a*) Heat the serum to 55°C. for half an hour ; (*b*) add 0·2 c.c. of the heated serum to 0·5 c.c. of guinea-pig's complement—incubate half an hour ; (*c*) add 0·5 c.c. of sheep's erythrocytes to (*b*)—incubate = complete hæmolysis.

It may be stated here incidentally that the above globulin-like substance does not inhibit the action of the natural complement normally present in the serum as long as it is not separated from the serum by the action of distilled water.

The anti-complementary globulin test, the globulin ring test, and the globulin precipitation test are of great diagnostic value in kala-azar. The last two tests may very rarely be present in some apparently obscure cases of enlarged spleen, in which no Leishman-Donovan bodies may be found on spleen puncture. But some of these may be cases of kala-azar, as they are cured by administration of urea stibamine, and as Leishman-Donovan bodies may not be found on spleen puncture in some rare cases of undoubted kala-azar (Leishman, Christophers).

The globulin-opacity test has already been described and, as stated before, being a quantitative one, gives more definite results than any other serum test. The *hæmolytic test*, the *aldehyde test* and its limitations have already been described. By taking a measured quantity of blood, say 20 c.mm., and placing it in 0·6 c.c. of distilled water in a small test-tube, Sia has noted the rate of sedimentation of the precipitate formed. If the solution becomes cloudy within five minutes the case is considered by him to be one of kala-azar. In advanced cases of kala-azar sedimentation is complete within fifteen minutes.

Chopra's Urea Stibamine Test.—Chopra and his co-workers have observed that when a solution of an organic compound of antimony is brought in contact with the serum from a kala-azar patient, a thick flocculent precipitate is formed at the junction of the two. The precipitate is much more marked and distinctive with urea stibamine than with other organic antimonials and, therefore, this is generally employed. It shows a tendency to conglomerate

to a mass at the junction of the two fluids and is difficult to break in by shaking, and does not dissolve if the tubes are allowed to stand for twenty-four hours or longer. As regards the strength of the solution required, it is found that although 0·05 per cent. or even weaker solutions give a precipitate, it is advantageous to use 1 to 4 per cent. solutions. Antimonyl tartrates do not give this precipitate. When normal sera or sera of patients suffering from other diseases are tested in the above way, there is either no precipitate whatever, or only a slight precipitate at the junction of the two fluids.

Technique.—The serum is pipetted out in a capillary tube and put into a miniature test-tube, 3 to 4 mm. in diameter. A 4-per-cent. solution of urea stibamine is slowly added along the side of the tube to form a layer on the top. With kala-azar serum a thick flocculent precipitate forms at once. The test has been further simplified by using the blood from a finger prick, so that it can be performed by the bedside. Two or three drops of blood from a finger prick are received in ¼ to ½ c.c. of a 2 per cent. solution of potassium oxalate (2 per cent. potassium citrate or normal saline may be used). A little of this is transferred by a capillary pipette into a glass tubing 3 to 4 mm. in diameter, sealed at one end. The solution of urea stibamine is then added, and this being heavier sinks to the bottom. Almost immediately a flocculent precipitate begins to form at the junction of the two fluids and begins to settle down in a characteristic manner, resembling a shower of rain, through the urea stibamine solution. In very early cases of kala-azar the precipitate may take a minute or two before it appears; but in well-developed cases it forms immediately and flocculation is quite distinct. The advantages claimed for the antimony test over the aldehyde test are: (*a*) It is given much earlier in the course of the disease than the aldehyde test. A definitely positive reaction is obtained from the fifteenth to twenty-first days of the disease. (*b*) The reaction is produced immediately, and it is not necessary to wait for twenty-four hours, as is sometimes the case with the aldehyde test. (*c*) The test can be performed with a much smaller quantity of serum.

Hodgson, Vardon and Singh's Test.—Hodgson, Vardon and Singh have described a preliminary note on a quick and simple test for the differentiation of malaria from kala-azar and other fevers. The technique is described as follows: "Prepare some sterile 5 per cent. citrate of soda, several narrow glass tubes about 5 mm. wide and 60 mm. long, marking them as follows from the bottom: 24 mm., 18 mm., 12 mm., 6 mm. Sterilize a 5 c.c. syringe and draw up into the syringe one part of 5 per cent. citrate of soda and four parts of patient's blood, mix and fill a glass tube up to the 24 mm. mark, allow mixture to stand for half an hour." When the blood of patients suffering from malaria was sedimented by the above method, the supernatant fluid was markedly yellow. In some malaria cases it was golden-yellow, others greenish-yellow, and a third group reddish-yellow. In proved cases of kala-azar there was a rapid clearing of the supernatant fluid, leaving it colourless or faint greenish-white.

For further details the reader is referred to the original paper by the author in the *Indian Journal of Medical Research*, January, 1927.

For other laboratory methods, see the Chapters on symptomatology, and diagnosis and differential diagnosis.

While fully endorsing the importance of laboratory methods, the author holds the view of Sir Leonard Rogers, that a large proportion of fevers in the tropics can be diagnosed by purely clinical methods.

II.

FORECAST.

We have described in the preceding pages the latest advances in the treatment of kala-azar by the use of urea stibamine and the campaign against it in Assam. The last word about its treatment has not yet been said. We hope that an antimonial compound will be discovered which it will be possible to administer orally, so that it may be used like quinine by the patients without their having to undergo the operation of intravenous injection, which is sometimes very difficult, especially in the case of children and patients with œdema. Such an advance will make the treatment and the campaign against the disease more easy and popular. We also hope that antimony analogues of salvarsan, neo-salvarsan and allied compounds will also be discovered. The day will come when, in studying the organic derivatives of antimony, one will be reminded of a quaint simile employed by Dr. Bertheim about the chemistry of organic arsenic compounds. "He compares it to a sleeping beauty slumbering, until quite recently, in an unfrequented corner of 'Beilstein,' but who, now awakened, appears as one of the fairy gifts which synthetic chemistry bestows from time to time upon mankind" (Morgan).

The economic effect of the discovery of the cure for kala-azar will be very great in future, especially in the provinces of Assam, Bengal, and in other parts of India where the disease occurs. A disease which was considered incurable for centuries, destroyed millions of human lives, ruined families, decimated villages and retarded prosperity in some parts of India, has now lost all its terrors.

This disease in its epidemic manifestation constituted, according to Rogers, the old "Burdwan fever" which raged in Bengal in the 'sixties and 'seventies, and converted many parts of Bengal into a "valley of the shadow of death." Its terrible nature is well described in the words of one of the writers on this epidemic fever: "The devastation of the epidemic has a very sad tale to tell. Countries that once smiled with peace, health, and prosperity, have been turned into hot-beds of disease, misery, and death. Villages that once rang with the cheerful, merry tune of healthful infants, now resound with loud wailings and lamentations. Huts, which offered too little space for their occupants, are left without a tenant. The skulls of human beings now strew the fields at every few yards' distance. The fell disease has mocked

every human effort, and absorbed in its powerful grasp, day by day and inch by inch, every blessed spot which once used to be prized for its salubrity."

General Gorgas, speaking in 1914 on yellow fever control, stated that its eradication would command the attention and the gratitude of the world and that the thing could be done. To-day yellow fever is in full retreat in the Americas. The same will one day be said of kala-azar, and it may be hoped that before long the disease will be completely banished from India and other parts of the world where it occurs. The signs of its retreat in Assam are already within sight, thanks to the intensive mass treatment of the disease with urea stibamine. When the disease disappears from the world, then one of the highest triumphs of tropical medicine will be achieved

III.

ADDENDUM.

A SHORT SUMMARY ON RECENT WORK ON THE TRANSMISSION PROBLEM OF KALA-AZAR IN CHINA, BY YOUNG AND HERTIG.

In a recent personal communication, Dr. Charles W. Young has very kindly furnished me with an account of the recent work on the subject carried out by him and Marshall Hertig in the Department of Medicine, Peking Union Medical College, Peking, China. The following is a summary of the work:—

Attempts to transmit kala-azar to experimental animals by means of fleas (Pulex, Xenopsylla, Ceratophyllus, and Ctenocephalus), *Cimex lectularius* and *C. pipistrelli*, and *Hematopinus* spp., from striped and giant hamsters, have been uniformly negative. In one case, however, in one pot in which infected and normal hamsters were present, the negative animal became positive in the known presence of *Cimex lectularius*.

There are marked discrepancies between the distribution of *P. major* var. *chinensis* and *P. sergenti*, and the distribution of kala-azar, and also in the behaviour of these two species in ways that would seem significant as regards the probability of their acting as transmitters of kala-azar. Thus, *P. major* re-feeds with reluctance, *P. sergenti* readily. *P. major* is moderately susceptible to infection after natural feeds on hamsters (56 per cent. of 195 attempts), *P. sergenti* markedly less so (2·4 per cent. of 661 attempts). When artificially fed, *P. major* yielded 83 per cent. out of 157 feeds, while *P. sergenti* gave 48 per cent. out of 349. In *P. sergenti* there is a definite tendency for the infection to die out after the fifth or sixth day. Though *P. sergenti* is the dominant sandfly in the kala-azar villages near Hsüchowfu, Kiangsu Province, yet it is infected with difficulty, and such infections tend to die out. *P. major*, on the other hand, while easily infected, may be very plentiful where there is

little or no kala-azar. Neither species has been infected by them following feeds on kala-azar patients, nor have infections with *Leishmania* been found in wild sandflies. Attempts to transmit the disease by means of sandflies have given negative results.

Naturally infected wild rodents have not been encountered in the endemic region in China, except in the case of one striped hamster.

Insects naturally infected with *Leishmania donovani* have not been found.

OBSERVATIONS ON CLASMATOCYTES IN EXPERIMENTAL KALA-AZAR.

In a recent personal communication, Dr. Hu, of China, has very kindly given me an account of Cash and Hu's work on clasmatocyte cells in experimental kala-azar in infected hamsters. They have observed that in these animals there is a specific relationship between the development of the clasmatocyte cells and infection with *Leishmania donovani*. In infected hamsters, they found a moderate degree of anæmia and leucopenia, and there was apparently an increased extra-medullary blood formation in the spleen.

The appearance of clasmatocytes in the blood is a striking phenomenon after infection. They found no such cells in the peripheral blood, and only 2·3 per cent. in the splenic blood in hamsters normally. In the case of infected hamsters their percentage rose to 18·8 in the peripheral blood and 19·4 in the splenic blood, four months after inoculation. In the case of bone-marrow they rose from 2·2 to 4·25 per cent.

Further work in this direction is necessary before any conclusions are arrived at.

PERIPHERAL LESIONS DUE TO *LEISHMANIA DONOVANI*.

We have already described certain skin lesions due to infection of the skin by *Leishmania donovani*. (See chapter on "Dermal Leishmanoid.")

Cash and Hu have found infection of the skin in a few cases of kala-azar with distinct increase of mononuclear cells about the blood-vessels. Many of these cellular forms in the subcutaneous tissue have been shown by them to be clasmatocytes by supravital staining with neutral red. They found that the parasites could be present in considerable numbers without causing any obvious change in the skin. None of their positive cases showed any discoloration of the skin, though several of those in whose skin no parasites were found showed moderate pigmentation, mainly of the face, arms and trunk. They consider that more acute and active cases must be studied before the importance of the skin and subcutaneous tissue, as foci from which the infection is naturally distributed, is established.

Young and Hertig have found the following peripheral lesions produced by *Leishmania donovani* and allied organisms in experimental animals.

Intraperitoneal inoculation of Acton and Knowles' "xanthoma" strain into Chinese striped hamsters (*Cricetulus griseus*) produced only visceral lesions. The same is true of cultures from a case of Brahmachari's "dermal leishmanoid."

The flagellates of both of these strains were agglutinated by the sera of rabbits immunized against Indian strains of *Leishmania donovani*. From these reactions it may be assumed that these organisms are *Leishmania donovani*.

These observers obtained from Nicolle, in Tunis, strains of the organisms from local human kala-azar, canine kala-azar and from the gecko, which had been under cultivation on NNN medium for many years. The cultures obtained from Tunis acted in a different and peculiar manner. They used strains of *Leishmania donovani* (*Leishmania infantum*, "kala-azar humain"); strains of *Leishmania canis* ("kala-azar canin"); and strains of *Leishmania tarentolæ* ("leptomonas de gecko") from the gecko. All of these strains originally produced visceral lesions only. When these cultures were inoculated intraperitoneally into hamsters, the infections were visceral at first, with enlarged spleen and liver and with leishmania fairly abundant in the smears from the spleen, liver, bone-marrow and heart blood. After a lapse of from two months to over a year from the time of inoculation, bilaterally symmetrical lesions began to appear in the following order: (1) swellings of the carpi and tarsi, extending later to the feet, including the digits; (2) swelling of the posterior half of the scrotum in males, with subsequent ulceration, infiltration and enlargement of the clitoris, exceptionally with ulceration of the perineum in the female; (3) swelling (infiltration) and later ulceration of the base of the tail; (4) similar swelling of the nose, rarely with ulceration; and (5) swelling and ulceration of the margins of the ears. From the swollen tissues leishmania enclosed in large mononuclear cells (clasmatocytes) were obtained, often in large numbers. The lesions of the feet never showed ulceration. The clasmatocytes were present often in enormous numbers between the connective-tissue fibres of the ligaments of the carpi and tarsi and distally feet, including digits. The cells containing parasites in the lesions showing ulceration were in the deep layers of the skin and subcutaneous tissues. Intraperitoneal inoculation of the tissues from these peripheral lesions produced the same picture in two to four months and continued to do so consistently and repeatedly. The same pathological changes were obtained with all the five strains used. As the peripheral lesions developed, those of the viscera tended to disappear, so that at autopsy some of the animals had normal-sized spleens and livers, negative for leishmania in smear. Intraperitoneal inoculation of such tissue did not cause infection in hamsters. Thus it will be seen that these strains which originally produced visceral lesions only, now caused pathological changes precisely like those described by Gonder, Laveran and Sergent for recently isolated *Leishmania tropica*. Similar lesions of a single extremity were found in two hamsters inoculated with two different Chinese strains of *Leishmania donovani*. These hamsters, already showing heavy visceral infection, had been tied out with leather thongs constricting the four extremities during insect feeding experiments several months before the appearance of the lesions. Inoculation of tissue from these local swellings produced only general visceral infections like those caused by the strains with which these hamsters had been originally infected.

TEMPERATURE IN RELATION TO CULTURE OF *LEISHMANIA DONOVANI*.

According to Christophers, Shortt and Barraud, the extreme range of temperature at which development was obtained was 16° C. and 34° C. Throughout this range the form of development appeared identical, the only variation observed being in the degree to which multiplication took place and the length of life of the culture or rapidity of development. Effective culture in which the parasites became very numerous was obtained between 22° C. and 30° C. Some difference in different strains was noticeable. Roughly speaking, some strains appeared to be more robust than others, and these, as a rule, appeared to culture over a wider range of temperature than others. At 28° C. no strain failed to culture freely. In some strains profuse culture was obtained at 30° C., and even at 32° C.; most strains, however, did not culture satisfactorily at these temperatures and many were negative. Existing cultures placed at 36° C. showed the flagellates at first in very active movement, but the cultures were usually killed, as shown by lack of motility and failure to subculture after about the third day. In spite of this, two positive cultures (from subcultures) were obtained at 34° C; these, however, quickly died out. Culture appeared to take place earlier at the high temperatures than at the lower ones; thus cultures at 28° C. were usually positive on the third day, whereas at 22° C., they did not usually show this condition until later.

Christophers, Shortt and Barraud hold that there would be nothing unexpected in the development of the parasite of kala-azar in the gut of the sandfly or elsewhere at a temperature of 28° C., or even at a higher temperature. If an early effective development is to be favoured, it is even probable that a temperature in this neighbourhood would be the most suitable, as tending to hasten development without being definitely hostile to it.

Though development of the flagellate form in culture can clearly take place through a wide range of temperature, it is evident that this range is pitched without any reference to the temperature of warm-blooded animals. The range is not, however, very divergent from that of the ordinary temperatures of external conditions in at least some tropical or subtropical countries.

PREPARATION OF THICK BLOOD-FILMS.

Some of the methods have already been described (see Appendix). Among others may be mentioned the following :—

Ruge's Method.—This is one of the best methods for the preparation of thick blood-films. After the blood has dried well, gently move the slide about in a glass containing a 2 per cent. solution of formalin to which has been added 1 per cent. glacial acetic acid. After laking is complete, treat the slide in the same way as above in a glass of tap water to remove all traces of acid. Next wash very gently in distilled water and stain.

The Method of James consists of making a circular smear about three quarters of an inch in diameter from a drop of blood and when dry it is treated with

acid alcohol consisting of a mixture of ten drops of HCl and 50 c.c. of absolute alcohol till dehæmoglobinization is complete. Then wash thoroughly in water for five to ten minutes. Stain after drying.

The Method of Shortt, Das and Lal.—They consider that thick film smears possess the disadvantage of presenting the white cells in a contracted condition or at least in a nearer approach to a globular form, which renders the search for parasites much more difficult, and often obscures their identity, as compared with the flattened-out white cells of a comparatively thin film.

A small drop of blood is placed at one end of a slide; a second slide is applied to it, as in making an ordinary blood-smear. The second slide, as soon as the blood has spread out along its edge, is pushed along the surface of the first with an even motion until the blood is almost exhausted. At this point, instead of continuing this motion, as in making an ordinary smear, the second slide is abruptly lifted off, with the result that the blood-smear ends in a straight edge stretching transversely across the slide. This straight edge is somewhat thicker than the rest of the smear and contains a large percentage of the total white cell content of the drop of blood. The white cells in the straight edge are all that it is necessary to examine for the purpose of determining, with a fair degree of accuracy, the presence and numbers of Leishman-Donovan bodies in the peripheral blood. As an invariable routine the terminal edges of four slides, prepared in the way described, are examined for parasites.

Shortt considers that the presence of a temperature above the normal has an effect in increasing the number of parasites in the peripheral circulation. At the same time he observes that many cases during long periods of apyrexia still show the presence of parasites in the peripheral blood. He considers that the activity of the disease has an effect on the parasite count, the periods of exacerbation as shown by raised temperature and all the concomitant factors of an active disease process increasing the parasite count in the blood. The parasites are found inside polymorphonuclear as well as mononuclear leucocytes.

The Method of Brahmachari and Sen.—Brachmachari and Sen have elaborated the following method of making and fixing thick film smears of blood.

A thin rectangular steel or tin or aluminium plate, about the size of an ordinary glass slide, is used as a blood-film spreader. One of the edges of the spreader is slightly notched in the middle, the notched portion being less than the breadth of the glass slide. Both the notched and unnotched portions must be even and absolutely parallel to the breadth of the spreader.

The film must not be very thick nor dried very slowly, otherwise the leucocytes become contracted and there is much difficulty in distinguishing Leishman-Donovan bodies inside them.

(1) A fairly large drop of blood is taken towards one end of a thoroughly clean glass slide and touched by the notched portion of the spreader, so that blood runs between the notched edge and the slide. Then the film is drawn, the spreader being held at an angle of 45°, the blood following it and not pushed before it. The thickness of the film may be varied by varying this

angle. Dry the slide in air or in a regulated vacuum desiccator, or in a 37° C. incubator. At room temperature it takes about half an hour to dry in the air. In the case of more thick films it may take one to two hours to dry in the air at ordinary temperatures.

(2) Put the slide in a glass tray containing acetone, "Merck," extra pure, for about five to ten minutes, according to the thickness of the blood-film, the tray being covered to prevent the acetone from evaporating during this period. Then allow the slide to dry till the acetone is evaporated.

(3) Dehæmoglobinize in distilled or tap-water. The process is finished within about one minute, and then wash with methyl alcohol to remove any further trace of acetone which may be left in the slide.

(4) Dry, and stain with one of the Romanowsky stains, such as Leishman or Wright.

The advantage of this method is that all structures of the blood, such as leucocytes, blood-platelets, &c., become fixed except the hæmoglobin of the erythrocytes, with the result that it can easily be washed away by the process of dehæmoglobinization.

More recently the authors have observed that if the blood is spread according to the method of Shortt, Das and Lal described above, then the result is also satisfactory.

TECHNIQUE FOR BRINGING OUT HISTOLOGIC STRUCTURE OF FLAGELLATES.

Parrot and Lestoquard's Method.—(*a*) Fix 10 min. in Schaudinn's alcoholic sublimate. (*b*) Transfer to iodine-alcohol. (*c*) Remove iodine with 95 per cent. alcohol and dry. (*d*) Immerse in horse-serum 10 min. and dry. (*e*) Stain with Giemsa.

REFERENCES.

1927 BRAHMACHARI, U. N., and SEN, P. B. A preliminary note on a new method of dehæmoglobinization of thick blood-films for the determination of malarial parasites and Leishman-Donovan bodies, etc., from the peripheral blood. *Cal. Med. Journ.*, vol. 22, No. 6, December, and *Ind. Journ. Med.*, vol. 8, Part 6, December.

1928 BRAHMACHARI, U. N., and SEN, P. R. On a new method of fixing thick blood-films for the finding of Leishman-Donovan bodies from the peripheral blood. *Cal. Med. Journ.*, February, and *Ind. Journ. Med.*, February. (In press.)

1927 CASH, J. R., and HU, C. H. Demonstration of *Leishmania donovani* in the skin and subcutaneous tissue of kala-azar patients. *Journ. Amer. Med. Assoc.*, vol. 89, No. 19, November 5, pp. 1576-7.

1927 CASH, J. R., and HU, C. H. The clasmatocyte in experimental kala-azar. (Under publication.)

1927 HU, C. H., and CASH, J. R. Considerations upon the relationship of the reticulo-endothelial system to kala-azar. *Proc. Soc. Exp. Biol. and Med.*, vol. 24, No. 6, March, pp. 469-472.

1927 SHORTT, H. E., DAS, S., and LAL, C. The finding of parasites in the peripheral blood of kala-azar cases by direct microscopical examination. *Ind. Journ. Med. Res.*, vol. 15, No. 2, October, pp. 529-538.

1927 YOUNG, CHARLES W., and HERTIG, MARSHALL. Kala-azar transmission experiments with Chinese sandflies (Phlebotomus). *Proc. Soc. Exp. Biol. and Med.*, vol. 24, pp. 823-825.

1927 YOUNG, CHARLES W. Recent research on the leishmaniases outside of China. *China Medical Journal*, vol. 41, No. 11, November, pp. 900-909.

The following papers were read at the Seventh Congress of the Far Eastern Association of Tropical Medicine, held in Calcutta, December 5 to 24, 1927 :—

Relation between Chemical Constitution of Antimonials and their Therapeutic Properties. (Section of Immunology and Chemotherapy.) By Dr. U. N. Brahmachari.

Action of Pentavalent Compounds of Antimony on the *Leishmania donovani* Parasites. By Major R. N. Chopra and Lt.-Col. Hugh W. Acton.

Observations on the Diagnostic Value of the Antimony Test for Kala-azar. By Major R. N. Chopra, Gupta and Basu.

The Kala-azar Transmission Problem and the Factor of Resistance. By Lt.-Col. R. Knowles.

Life-History of *Leishmania donovani* in its Insect and Mammalian Hosts. By Major H. E. Shortt.

Peripheral Lesions produced by *Leishmania donovani* and Allied Organisms. By Charles W. Young and Marshall Hertig.

The Presence of *Leishmania donovani* in the Skin and Subcutaneous Tissue in Cases of Kala-azar. By J. R. Cash and C. H. Hu.

Kala-azar Studies in North China. By Charles W. Young and Marshall Hertig.

Bibliography.

The literature on the subject of leishmaniasis is now very extensive. Attempts have been made here to make the subject of human kala-azar (or visceral leishmaniasis) complete, and only a few references to oriental sore and canine leishmaniasis have been given.

1911 ABATE, A. Ricerche ematologiche nella leishmaniosi infantile. Gazetta Internazionale, 1911, No. 41, p. 972. Ref. in Pathologica, 1912, June 15, vol. 4, No. 87, p. 302.

1913 ——— La resistanza dei globuli Rossi nelle leishmaniosi infantile. Malaria e Malat. d. Paesi Caldi, June July, vol. 4, No. 4, p. 263.

1914 —— Ulteriore contributo alla casistica della leishmaniosi. Lavori d. Soc. Italiana di Patalogia, Esotica, pp. 68-74.

1917 —— Contributo all' ematologia del kala-azar infantile. Malaria et Malat. d. Paesi Caldi, April 20, vol. 8, No. 2, pp. 68-76.

1917 ——— Il kala azar infantile a Catania (Note epidemiologiche e nosografiche). Malaria e Malat. d. Paesi Caldi, April 20, vol. 8, No. 2, pp. 57-68.

1927 ACTON, H. W., and NAPIER, L. E. Post-kala-azar dermal leishmaniasis. Ind. Journ. Med. Res., vol. 15, No. 1, July, pp. 97 106.

1925 ACUNA ET AL. Kala-azar in children. Prensa Med., Buenos Aires, 1924, Nov. 30, vol. 2, p. 585. Summarized in Journ. Amer. Med. Assoc., Jan. 17, vol. 84, No. 3, p. 237.

1926 ADAMPOULOS, CH. Le kala-azar dans le départment de Messénie. Grèce Méd., July-August, vol. 28, Nos. 7 8, p. 26.

1924 ADELHEIM, R. Über leishmaniosis infantum et canina in Riga. Arch. für Schiffs- u. Tropen-Hyg., vol 28, No. 9, pp. 367-387 (Ref. in Lancet, Dec. 6, p. 1190).

1921 ADIE, H. A. Preliminary note on the development of the Leishman-Donovan parasite in spleen juice and in the alimentary tract of *Cimex lectularius* (Linn.). Ind. Journ. Med. Res., Oct., vol. 9, No. 2, pp. 255-260.

1922 ——— A note on bodies observed in *Cimex rotundatus* Linn., collected in a kala-azar-infected area in Assam. Ind. Journ. Med. Res., vol. 10, July, No. 1, pp. 230-238.

1922 —— Telegram announcing finding Leishman-Donovan bodies in salivary glands and ducts of *Cimex rotundatus*. Ind. Journ. Med. Res., January, vol. 9, No. 3, p.v.

1927 ADLER, S., and THEODOR, O. The behaviour of cultures of *Leishmania tropica*, *L. infantum*, and *L. brazilicuse* in the sand-fly, *Phlebotomus papatasii*. (Correspondence.) Nature, vol. 119, pp. 48-49.

1927 ——— ——— The behaviour of cultures of *Leishmania* sp. in *Phlebotomus papatasii*. Nature, vol. 119, p. 565.

1927 AGRONICK, M. A. Beiträge zur Epidemiologie und Kasuistik der Orientbeule. Dermat. Wochenschr., vol. 84, pp. 261-273.

1926 AGUIAR PUPO, J. Amino-arseno-phenol therapy. Compt. Rend. Soc. de Biol., vol. 95, pp. 993-994.

1905 AIRDIE. Leishman body found in China (Hankow). Journ. of Trop. Med. and Hyg., vol. 8, p. 220.

1923 AKAMATSU (TOKUJIRO). On kala-azar in Shantung district in China. Jikken Igaku Zasshi (Journ. Experim. Med.), Dec., vol. 7, No. 11-12.

1921 ALLEN & HANBURYS. Intravenous injection of stibenyl in kala-azar. Lancet (Correspondence), May 21, p. 1102.

1910 ALVARES, DIONYSIO. Um caso de kala-azar infantil em Lisboa. Sociedade das Sciencias medicas de Lisboa, Sessao de 12 de Marco de 1910. A Medicina Contemporanea, Lisboa, March 20, vol. 28, pp. 90-91.

1910 —— and DA SILVA. Sobre a existencia do kala-azar espontaneo no cao em Lisboa. Sobre a frequencia do kala-azar nos caes em Lisboa. A Medicina contemporanea, May 22, vol. 28, p. 162.

1911 ——— ——— Um novo caso de kala-azar em Portugal. A Medicina contemporanea, vol. 29, p. 5.

1911 —— Sobre a presenca de formas de leishmania na pulga. A Medicina contemporanea, May 20, vol. 28, p. 197.

1926 ANDERSON, C. Leishmaniasis in North Africa. Rev. Tunisienne Sci. Med., March, 8 pp.

1922 to 1927 ANNUAL REPORTS OF THE CALCUTTA SCHOOL OF TROPICAL MEDICINE.

1911 ANNUAL REPORT OF THE SANITARY COMMISSIONER WITH THE GOVERNMENT OF INDIA for 1910, with appendices and returns of sickness and mortality among European troops, native troops, and prisoners in India for the year. Folio. Calcutta: Superintendent, Government Printing, India. Kala-azar and oriental sore, pp. 49, 119-120.

1927 ARAGÃO, H. Palestra sobre leishmanioses. Sciencia Med., vol. 5, pp. 121-132.

1916 ARAVANDINOS, A. Beobachtungen über die innere leishmaniosis in Griechenland. Arch. f. Schiffs- u. Trop.-Hyg., April, vol. 20, No. 8, pp. 193-203.

1916 —— L'anatomia patalogica e l'istopatologia di un caso greco di leishmaniosi interna. Malaria e Malat. d. Paesi Caldi, April 20, vol. 7, No. 79-82.

1916 —— Contribution à l'historique de la leishmaniose interne. Bull. Soc. Path. Exot., Jan., vol. 9, No. 1, pp. 10-13.

1916 —— Modification dans la technique de la ponction de la rate. Bull. Soc. Path. Exot., July, vol. 9, No. 7, pp. 444-448.

1918 —— Méthode pour assurer l'innocuité parfaite de la ponction splénique. Bull. Soc. Path. Exot., October 9, pp. 701-715.

1911 —— and MICHAELIDES, NICOL. Kala-azar in Griechenland. I. Das kala-azar auf der Insel Hydra. Zentralbl. für innere Med., Leipzig, April 15, vol. 32, No. 15, pp. 360-375.

1907 ARCHER, G. J. S. A case of kala azar contracted in Crete. Journ. Roy. Army Med. Corps, vol. 9, p. 287, *bis* 290.

1907 —— Notes on the post-mortem examination of a case of kala-azar contracted in Crete. Journ. Roy. Army Med. Corps, vol. 9, pp. 511-513.

1910 ARCHIBALD, R. G. The alkalinity of the blood serum in kala-azar. Journ. Roy. Army Med. Corps, June, vol. 14, No. 6, pp. 615-620.

1911 —— The alkalinity of the blood serum in kala-azar. Fourth Report of the Wellcome Tropical Research Laboratories at the Gordon Memorial College, Khartoum. Vol. A.—Medical, pp. 173-177.

1913 —— An interesting case of kala-azar. Journ. Roy. Army Med. Corps, May, vol. 20, No. 5, pp. 512-521.

1914 —— A preliminary report on some further investigations on kala-azar in the Sudan. Journ. Roy. Army Med. Corps, Nov., vol. 23, No. 5, pp. 479-495.

1922 —— The leishmaniasis. The practice of medicine in the tropics, edited by W. Byam and R. G. Archibald.

1923 —— Kala-azar in the Sudan, with special reference to its treatment by tartar emetic. Amer. Journ. Trop. Med., July, vol. III, No. 4, pp. 307-324

1924 —— and SOSEL, B. A sporozoon from the spleen of a case of splenomegaly in the Sudan. Trans. Roy. Soc. Trop. Med. and Hyg., vol. 17, No. 8, Feb. 21, pp. 482-484.

1923 ARCHIV. DEL INSTITUTO NACIONAL DE HIG. DE ALFONSO XIII. August, vol. 2, No. 2, pp. 67-84. Nuevos cas de kala-azar infantile en el centro de Espana (Caceres, Madrid, Toledo), un caso de muerte por el stibenyl.

1914 ARCHIVES DE L'INSTITUT PASTEUR DE TUNIS. Aug. 1, vol. 9, No. 1, pp. 30-38. Chronique du Kala-azar en Tunisie.

1924 ARCOLEO, GAETANO. Ein Fall von Kala azar bei einem Muselmann, der mit "Beyer 205" geheilt wurde. Arch. f. Schiffs u. Tropen-Hyg., August, vol. 28, No. 7, pp. 295-297.

1925 ARENA, G. Sull' importanza diagnostica della reazione di Brahmachari nella diagnosi di kala-azar. Clin. pediat. Modena, vol. 7, pp. 161-169.

1927 —— Contributo allo studio della patogenesi della leucopenia nel kala-azar infantile (The pathogeny of leucopenia in infantile kala-azar). Pediatria, May 1, vol. 35, No. 9, pp. 465-475.

1926 ARIAS ARANDA, C. Case in Salta. Bol. Inst. de Clin. Quir., vol. 2, pp. 322-326.

1926 ARTAMONOV, A. S. Kala-azar at Samarchand. Treatment by stibenyl. Russ. Journ. Trop. Med., Nos. 6-7, pp. 35-40. (In Russian.)

1927 —— The distribution of kala-azar in Samarkand and its cure by antimony preparations. Arch. f. Schiffs- u. Trop.-Hyg., January, vol. 31, No. 1, pp. 32-37.

1927 —— Zur Frage der Verbreitung der innerlichen Leishmaniasis in Samarkand und die Heilung der Krankheit durch die Präparate des Stibiums. Arch. f. Schiffs- u. Tropen-Hyg., vol. 31, pp 32-37.

1904 ASCHOFF, LUDWIG. Demonstration von präparaten eines Falles von kala-azar. Verhandlungen der deutschen Pathologischen Gesellschaft, vol. 7, pp. 81-82.
1904 —— Demonstration eines Falles von Kala-azar. Zentrabl. für allgemeine Pathologie und pathologische Anatomie, vol. 15, p. 537.
1910 ASPLAND, W. G. Is ponos kala-azar? Brit. Med. Journ., Jan. 15, p. 139.
1906 AUER, JOHN. Some hitherto undescribed structures found in the large lymphocytes of a case of acute leukæmia. Amer. Journ. of Med. Sci., vol. 131, pp. 1002-1015. Comparison with Leishman-Donovan bodies, p. 1014.
1924 AURICCHIO, LUIGI. Ricerche sulla glicemia nella leishmaniosi infantile. Pediatria, June 15, vol. 32, No 12, pp. 704-711.
1927 —— Considerazioni e ricerche sulla terapia della leishmaniosi infantile (Treatment of infantile leishmaniasis). Pediatria, March 15, vol. 35, No. 6, pp. 289-300.
1924 AVARI, C. R., & MACKIE, J. P. Canine leishmaniasis in Bombay. Ind. Med. Gaz., Dec., vol. 59, No. 12, pp. 604-605.
1922 AWATI, P. R. Survey of biting insects of Assam with reference to kala-azar for the whole year from November, 1921, to October, 1922. Ind. Journ. Med. Res., vol. 10, No. 2, pp. 579-591.
1922 AYYAR, T. S., & KRISHNAN, K. V. The value of culture of the peripheral blood in kala azar as a diagnostic procedure. Ind. Med. Gazette, July, vol. 57, No. 7, pp. 255-256.

1909 BABINGTON, M. H. Notes on a case of a disease prevalent in Malta and known there as splenic leucocythæmia. Journ. Roy. Army Med. Corps, Sept., vol. 13, No. 3, pp. 291-294.
1910 —— Case of kala-azar in Malta. Trans. United Services Medical Society, October December. Abstract in Journ. Trop. Med. and Hyg., 1911, vol. 14, No. 8, p. 125.
1911 —— Case of kala-azar. Journ. Roy. Army Med. Corps, Oct., vol. 17, No. 4, pp. 380-386.
1924 BAGCHI, H. N. A case of kala-azar, having intolerance towards sodium antimonyl tartrate cured by urea stibamine. Ind. Journ. Med., vol. 5, pp. 145-147.
1910 BAKER, W. L. Some notes on a case of kala-azar in Malta. Trans. United Services Medical Society. October-December. Abstract in Journ. of Trop. Med. and Hyg., 1911, vol. 14, No. 8, p. 125.
1911 —— Some notes on a case of kala-azar in Malta. Journ. Roy. Army Med. Corps, October, vol. 17, No. 4, p. 380.
1925 BALASUBRAMANIAN, T. S. Kala azar in the Ramnad District of the Madras Presidency. Ind. Journ. Med. Res., July, vol. 13, pp. 105-106.
1920 BALFOUR, ANDREW. Kala-azar in Mesopotamia. Brit. Med. Journ., Aug. 28, p. 335.
1908 —— & ARCHIBALD, R. G. Review of some of the recent advances in tropical medicine, hygiene, and tropical veterinary science, with special reference to their possible bearing on medical, sanitary and veterinary work in the Anglo-Egyptian Sudan. Being a Supplement to the Third Report of the Wellcome Research Laboratories at the Gordon Memorial College, Khartoum. (Leishmaniasis, pp. 95-99.)
1911 —— —— —— Case of kala-azar treated with "606." Fourth Report of the Wellcome Tropical Research Laboratories at the Gordon Memorial College, Khartoum. Vol. A. Medical, p. 185, *bis* 190.
1911 —— —— in collaboration with FRY, Capt. W. B., & O'FARRELL, Capt. W. R. Leishmaniasis, pp. 150-157. A Supplement to the Fourth Report of the Wellcome Tropical Research Laboratories at the Gordon Memorial College, Khartoum.
1910 BALSAMO, G. Sul kala azar in Calabria. Studi intorno ad alcune malattie tropicali in Sicilia e Calabria. Tipografia Labicana, vol. 1, p. 41.
1913 BANDI, IVO. Preliminary note on the identity of certain leishmaniases based on biological reactions. Journ. Trop. Med. and Hyg., Feb. 15, vol. 16, No. 4, p. 50.
1922 BANERJEE, D. N. An announcement. A preliminary note on the diagnosis of kala azar by examination of bone marrow. Cal. Med. Journ., April, vol. 16, No. 10, p. 1.
1923 —— Some interesting kala-azar cases. Cal. Med. Journ., June, vol. 17, No. 12, pp. 265-267.
1923 —— Cultivation of *Leishmania donovani*. Cal. Med. Journ., Sept., vol. 18, No. 3, pp. 417-420.
1923 —— Field work for control of kala-azar in Bengal. Ind. Med. Record, Aug., pp. 223-226, and Sept., pp. 241-243.

1923 BANERJEE, D. N. Observations on the occurrence of leishmania in ulcers of stomach in a case of kala azar. Cal. Med. Journ., Sept., vol. 18, No. 3, pp. 385-388.
1923 —— Kala-azar. Cal. Med. Journ., January, vol. 17, No. 7, pp. 7-42.
1924 —— The latent phase of kala-azar. Cal. Med. Journ., March, vol. 19, No. 9, pp. 655-658.
1924 —— A scheme for the control of kala-azar in Bengal. Ind. Med. Rec., Feb., vol. 43, No. 2, pp. 36-38.
1923 —— & SHAH, J. C. A study of blood sugar in Bengalees in health and in kala-azar. Cal. Med. Journ., vol. 17, March, No. 9, pp. 109-114.
1923 —— —— On the possible functions of adrenalin in kala azar. Cal. Med. Journ., April, vol. 17, No. 10, pp. 145-150.
1923 BANERJI, J. Organisation of kala azar centres in Bengal. Ind. Med. Rec., Aug., pp. 227-229.
1926 BARBACCI, P. Sur un cas leishmania infantum observé à Sienne (Italie). Remarques épidémiologiques. Bull. Soc. Path. Exot., vol. 19, pp. 11-14.
1927 —— Un secondo caso di leishmaniosi interna infantile osservato in Siena. Policlin., sez. prat., vol. 34, pp 667-671.
1924 BARBERI, S. Restauro dei proteici del plasma nel kala azar. Pediatria, April 1, vol. 32, No. 7, pp. 399-407.
1927 BARNARDO, F. A. F. Difficulties in the early diagnosis of the typhoid group of fevers. Ind. Med. Gazette, July, vol. 62, No. 7, pp. 393-400.
1926 BARRAUD, P. J. Report upon a sandfly survey of Madras Town. Ind. Journ. Med. Res. Memoir, No. 4, pp. 207-218.
1917 BARRIO, N. GONZALEZ. Estudios sobre la anatomia patologica del kala azar infantil. Bol. Inst. Nac. Higien. de Alfonso XIII, June 30, vol. 13, No. 50, pp. 117-130.
1917 —— Estudios sobre la anatomia patologica del kala azar infantil. Bol. Inst. Nac. Higien. de Alfonso XIII, Sept. 30, vol. 13, No. 51, pp. 172-178.
1925 —— Nuevos casos de kala-azar en Madrid y su provincia; resultados del tratamiento. Arch. de cardiol. y hematol., vol. 6, pp. 200-205.
1910 BASILE, C. Sulla leishmaniosi dei cane e sull' ospite intermedio del kala azar infantile. Rend. R. Accad. dei Lincei Rome, vol. 19, pp. 523-527.
1910 —— Alcune osservazione sulla presenza di leishmanie nei cani. Rend. R. Accad. dei Lincei, Rome, vol. 19, pp. 158-160.
1911 —— Sulla leishmaniosi e sul suo modo di trasmissione. Rend. R. Accad. dei Lincei, Rome, Feb. 19, 5th serie, vol. 20, pp. 278-282, and Rend. R. Accad. dei Lincei, Rome, March 19, 5th serie, vol. 20, pp. 479-485.
1911 —— Sulla trasmissione della leishmaniosi. (Nota preventiva.) Archivo trimestrale redatto du Umberto Gabbi. pp. 6-8, consecutive numbering, pp. 46-48.
1911 —— Sulla trasmissione della leishmanioso. Rend. R. Accad. dei Lincei, Rome, January, 8, vol. 20, pp. 50-51.
1911 —— Sulla leishmaniosi e sul suo modo di trasmissione. Gazzetta degli Ospedali e delle Cliniche, pp. 372-373.
1911 —— Sulla leishmaniosi e sul suo modo di trasmissione. Nota 6a preliminare. Atti della reale Accademia dei Lincei, Rendiconti, Rome, 18 giugno, 5 serie, vol. 20, 1° semestre, fasc. 12, pp. 955-959.
1911 —— Sulla leishmaniosi e sul suo modo di trasmissione. Aggiunta alla 6a nota preliminare. Atti della reale Accademia dei Lincei, Rendiconti, Rome, 16 luglio, 5 serie, vol. 20, 2° semestre, fasc. 2, pp. 72-73.
1912 —— Sur l'identité des leishmanioses et sur leur mode de transmission. Bull. Soc. Path Exot., Dec., vol. 5, No. 10, pp. 812-816.
1912 —— Sull' identità e sul modo di trasmissione delle leishmaniosi. Contributo agli studi clinici, sperimentali ed epidemiologici. Il Policlinico, sezione medica, April, vol. 19, pp. 165-167.
1913 —— La trasmissione sperimentale delle leishmaniosi del Mediterranee ai topi conigli e cavie. Atti della Reale Accad. d. Lincei, Rendiconti, March 16, vol. 22 (1 Semest), No. 6, pp. 302-303.
1913 —— I recenti studi sull' identità della leishmaniosi umana e canina de Mediterraneo. Policlinico sez. Pratica, July 20, vol. 20, No. 29, pp. 1029-1032.
1913 —— Sulla leishmaniosi nel cani e sull' esistanza di leishmania nel midollo spinale di cani naturalmente infetti. Atti della Reale Accad. de Lincei, Rend., April 20, vol. 22 (1 semest.), No. 8, pp. 524-527.
1913 —— La trasmissione sperimentale delle leishmaniosi del Mediterranee ai topi per mezzo delle Pulci. Atti della Reale Accad. d. Lincei, Rendiconti, April 6, vol. 22 (1 Semest.), No. 7, pp. 468-470.
1913 —— Su alcuni ricerche etiologiche in un caso di leishmaniosi del Mediterraneo lettere all' Editore) Pathologica, July 15, vol. 5, No. 113, pp. 447-448.

1914 —— La meteorologia della leishmaniosi interna nel Mediterraneo. Nota 1. Atti d. R. Accad. d. Lincei, Rendiconti, series 5, April 5, vol. 23 (1 semest.), No. 7, pp. 539-542.
1914 —— La meteorologia della leishmaniosi interna nel Mediterraneo. Nota 2. Contributo critico agli Esperimenti di Trasmissione. Atti d. R. Accad. d. Lincei, Rendiconti, series 5, April 19, vol. 23 (1 semestre), No. 8, pp. 625-629.
1916 —— Leishmaniosi interna. Annali d'Igiene, April 30, vol. 26, No. 4, pp. 248-268.
1920 —— Leishmania, herpetomonas and crithidia in fleas. Parasitology, Dec., vol. 12, No. 4, pp. 366-377.
1920 —— La transmissione sperimentale delle leishmaniosi del Mediterraneo ai topi per mezzo delle Pulci.
1911 —— and VISENTINI. Sull' identita della leishmaniosi. Rendi. R. Accad. dei Lincei, Rome. April 23, 5th serie, vol. 20, pp. 590-591.
1911 —— LA CAVA, F., and VISENTINI, A. Sulla identità delle leishmaniose. (Studio particolareggiato delle condizioni di ambiente in cui si inizio e si svolse un caso di kala-azar). Nota seconda preliminaire. Atti della reale Accad. dei Lincei, Rendiconti, Rome, August 6, 5 serie, vol. 20 (2 semestre), Fasc. 3, pp. 150-154.
1911 —— —— —— Sopra un caso di leptomeningite da leishmania. Atti della reale Accademia dei Lincei, Rendiconti, July 16, 5 serie, vol. 20 (2 semestre); vol. 2, pp. 69-72.
1903 BASSETT-SMITH, P. W. The relationship of kala-azar with Mediterranean fever, and some details of the hæmatology of the latter. Journ. of Trop. Med. and Hyg., Feb. 2, vol. 6, pp. 37-30.
1905 —— Bruhl's disease, with special reference to the blood changes found, and connection with the Leishman-Donovan bodies. Brit. Med. Journ., vol. ii, pp. 1260-1261.
1907 —— Kala-azar in the Royal Navy. Transactions of the Soc. of Trop. Med. and Hyg., 1907-8, vol. I, pp. 121 125.
1908 —— Kala-azar in the Royal Navy. Journ. of Trop. Med. and Hyg., March 16, pp. 85-86.
1908 —— Kala-azar in the Royal Navy; with illustrative cases. Brit. Med. Journ., vol. 1, pp. 1043-1044.
1909 —— Kala azar in the Far East. Brit. Med. Journ., Dec. 4, vol. 2, p. 1614.
1913 —— Kala-azar in an adult from Malta. Journ. of Roy. Army Med. Corps, Nov., vol. 21, No. 5, pp. 581-584.
1914 —— Case of kala-azar (parasitic splenomegaly) in an adult from Malta. Proc. Roy. Soc. Med., March, vol. 7, No. 5 (Clin. Sect.), pp. 87-90.
1914 —— Discussion on kala-azar or parasitic splenomegaly and allied infections. Brit. Med. Journ., pp. 1058-1060.
1923 —— Naval medical history of the war. Section of tropical and sub-tropical medicine. Part II, Kala-azar. Journ. Roy. Nav. Med. Ser., July, vol. 9, No. 3, pp. 201-203.
1921 BASU, C. C. Observations on the chemical behaviour of malarial pigment, with a note on the nature of pigment found in the liver of kala-azar cases. Ind. Journ. Med., March, vol. 2, No. 1, pp. 328-331.
1927 —— A silver method of staining *Leishmania donovani* in the tissues. Ind. Med. Gaz., May, pp. 253 254.
1923 BASU, U. P. My experience of kala-azar and its treatment by antimony injections. Ind. Med. Record, Aug., pp. 229-230.
1924 BASU, S. C. Age incidence of kala-azar patients. Ind. Med. Record, vol. 43, No. 4, pp. 107-108.
1924 —— A note on an epidemic of kala-azar with typhoid-like onset and simultaneous occurrence in several families in a village in Bengal, including short reports of a few apparently similar epidemics of fever in some other villages. Calcutta Med. Journ., August, vol. 19, No. 2, pp. 58-72.
1924 BASU, INDU BHUSAN. Analysis of 50 clinically-diagnosed kala-azar cases treated in the out-patient department of Carmichael Medical College Hospitals. Calcutta Med. Journ., May, vol. 18, No. 11, pp. 745-752.
1922 —— Chronic malaria and kala-azar. Cal. Med. Journ., October, vol. 17, No. 4, pp. 153-169.
1912 BAYON, H. Demonstrations of specimens relating to the transmission of artificial cultures of *Leishmania infantum* to mice and rats. Brit. Med. Journ., Nov. 2, pp. 1197-1199.
1923 BECKER, E. R. Studies on the relationship between insect flagellates and leishmania. Amer. Journ. Hyg., July, vol. 3, No. 4, pp. 462-468.
1919 BELL, H. W. A case of kala-azar treated with tartar emetic. China Med. Journ., January, vol. 33, No. 1, pp. 13-14.

1902 BENTLEY, CHARLES A. Epidemic Malta fever in Assam. A short preliminary notice of certain recent discoveries relating to the true nature of kala-azar. Ind. Med. Gazette, vol. 37, pp. 337-339.
1902 —— Malaria and kala-azar. Ind. Med. Gazette, vol. 37, pp. 450-465.
1902 —— Kala-azar as an analogous disease to Malta fever. Journ. of Trop. Med. and Hyg., Sept. 1, p. 270.
1902 —— Kala-azar as an analogous disease to Malta fever: preliminary notes of an investigation and some discoveries regarding the nature of the condition known as kala-azar. Brit. Med. Journ., vol. 2, pp. 872-879; Journ. of Trop. Med. and Hyg., Jan. 1, 1903, vol. 6, pp. 8-16.
1904 —— Some notes on kala-azar and the new parasite. Journ. Trop. Med. and Hyg., vol. 7, p. 261.
1904 —— A short note on the parasite of kala-azar. Journ. Roy. Army Med. Corps, vol. 2, pp. 747-749.
1904 —— A short note on the parasite of kala-azar. Ind. Med. Gazette, March, vol. 39, pp. 81-82.
1904 —— Notes upon kala-azar and the new parasite. Brit. Med. Journ., Sept. 17, vol. 2, pp. 653-655.
1926 BERTRAND, M. New Algerian cases of kala-azar. Arch. Inst. Pasteur d'Algérie, vol. 4, p. 30.
1922 BESSEMANS, A. Influence de la concentration des sérums sur leur formolgelification et sur leur pouvoir formolgelifiant. Influence de la température sur leur formolgelification. C. R. Soc. Biol., July 1, vol. 87, No. 24, pp. 398-401.
1922 —— Influence de la dilution sur le pouvoir formolgelifiant des sérums. C. R. Soc. Biol., pp. 401-404.
1925 BHADURI, B. N. Retinal hæmorrhage in kala azar. Calcutta Med. Journ., 1925-26, vol. 20, pp. 186-192.
1927 —— Retinal hæmorrhage in kala azar. Cal. Med. Journ., January, pp. 327-341.
1927 —— Urea stibamine amblyopia. Cal. Med. Journ., January, vol. 21, No. 7, pp. 327-341.
1927 —— A case of opacities of the lens occurring as a sequel to an attack of kala-azar. Ind. Med. Gazette, March, vol. 62, No. 3, p. 144.
1927 BHATTACHARYA, S. C. An interesting case of kala-azar. Med. Rev. of Reviews, vol. 2, pp. 124-125.
1906 BIRT, C. The Leishman body, the gregarine stage of a herpetomonas. Journ. Roy. Army Med. Corps, vol. 6, pp. 653-658.
1906 —— and BATEMAN, H. R. Kala azar. Journ. Roy. Army Med. Corps, vol. 7, pp. 341-347.
1923 BIZARD, E., and TERRIEN, E. Cas de Leishmaniose interne chez une adulte contracté en France. Bull. Soc. Path. Exot., Feb. 14, vol. 16, No. 2, pp. 80-91.
1923 BLACKLOCK, B. The ætiology of kala-azar and tropical sore. Lancet, August 11, pp. 273-274.
1904 BLACKWELL, C. T. Report of a case of Dum-dum fever. With note by Major W. B. Leishman, R.A.M.C. Journ. Roy. Army Med. Corps, March, vol. 2, pp. 313-319.
1904 BLANCHARD, R. Note critique sur les corpuscles de Leishman. Revue de Médecine et d'Hygiène tropicales, vol. 1, pp. 37-42.
1901 BODDAERT, A. Contribution à l'étude du Ponos, d'après le texte hellène du docteur C. Jeannacopoulos. Annales de la Société de Médecine de Gand, vol. 80, 7e livr., pp. 315-319.
1926 BOISSEAU, A. Case of kala azar contracted in S. America. Arch. Inst. Pasteur d'Algerie, vol. 4, pp. 31-34.
1912 BOSE, K. C. The relation of kala-azar to malaria. Proc. 3rd Meet. Gen. Mal. Com. (Madras), Nos. 18-20, pp. 267-270.
1908 BOSU, B. B. Kala-azar in Patna. Ind. Med. Gazette, vol. 43, p. 210.
1926 BOUCHE, B. J. A case of kala-azar in the Simla Hills. Ind. Med. Gazette, vol. 61, pp. 20-21.
1916 BOUILLIEZ, M. Un cas de kala azar infantile au Moyen-Chari (Territoire du Tchad). Bull. Soc. Path. Exot. May, vol. 9, No. 5, pp. 290-302.
1908 BOUSFIELD, L. Observations on kala-azar in Kassala Province. Third report of the Wellcome Research Laboratories at the Gordon Memorial College, Khartoum, vol. 3, pp. 107-119.
1910 —— A tour of investigation as to prevalence of kala-azar in Kassala and Blue Nile districts, Sudan, from January 12 to May 16, 1909. Journ. Roy. Army Med. Corps, vol. 15, No. 2, pp. 161-183 and No. 3, pp. 292-307.

1911 Remarks on kala-azar in the Kassala and Blue Nile districts of the Sudan. Fourth Report of the Wellcome Tropical Research Laboratories at the Gordon Memorial College, Khartoum, vol. A., pp. 127-141.
1912 —— Some remarks on kala-azar in the Sudan. Transactions of the Soc. of Trop. Med. and Hyg., April, vol. 5, No. 6, pp. 234-239.
1906 BRAHMACHARI, U. N. A contribution to the study of fevers due to Leishman-Donovan bodies. Calcutta Med. Journ., October.
1906 —— Forms of pyrexia due to Leishman-Donovan bodies. Ind. Med. Gazette, January, vol. 41, p. 15.
1908 —— Sporadic kala-azar in Calcutta with notes of a case treated with atoxyl. Brit. Med. Journ., May 30, vol. 1, pp. 1286-1288.
1908 —— Fatty liver in kala-azar. Brit. Med. Journ., Sept. 26, vol. 11, p. 876.
1909 —— On the relative hæmoglobin-value of the resistant erythrocytes during the hæmolysis of blood and on the permeability of the erythrocytes to water as a factor in the production of hæmolysis. Bio-Chemical Journ., vol. 4, Nos. 5, 6 and 7.
1911 —— On the nature of the epidemic fever in Lower Bengal commonly known as Burdwan fever (1854-55). Ind. Med. Gazette, vol. 46, Sept., No. 9, 340-343.
1915 —— A preliminary report on the treatment of kala-azar with intravenous injection of metallic antimony. Ind. Med. Gazette, vol. 50, No. 12, pp. 455-457.
1916 —— Preparation of stable colloidal antimony. Lancet, October 21.
1916 —— Further observations on the treatment of kala-azar with metallic antimony and reports of cases treated with galyl, formaldehyde and Plimmer's salt. Ind. Med. Gazette, vol. 51, No. 1, pp. 16-18.
1916 —— Colloids and other drugs in the treatment of kala-azar. Calcutta Med. Journ., September, vol. 12, No. 3, pp. 70-72.
1916 —— Third report on the treatment of kala-azar with special reference to the use of antimony and formaldehyde. Ind. Med. Gazette, May, vol. 51, No. 5, pp. 173-178.
1917 —— Fourth report on the treatment of kala-azar and some blood reactions in this disease. Ind. Med. Gazette, Sept., vol. 52, No. 9, pp. 319-322.
1917 —— On the presence of an easily precipitable anticomplementary globulin-like substance in human serum and its importance in the diagnosis of kala-azar. Ind. Med. Gazette, Dec., vol. 52, No. 12, pp. 429-431.
1917 —— Kala azar, its treatment. Butterworth and Co., 1st Edition.
1920 —— Kala-azar, its treatment. Butterworth and Co., 2nd Edition.
1920 —— Treatment of kala azar with intramuscular injection of hyperacid antimonyl tartrate | urethane. Ind. Med. Gazette, May, vol. 55, No. 5, pp. 176-177.
1921 —— The treatment of kala-azar with some new antimonial preparations. Journ. Trop. Med. and Hyg., August 15, vol. 24, No. 16, pp. 213-215.
1922 —— A new form of cutaneous leishmaniosis. Dermal leishmanoid. Ind. Med. Gazette, April, vol. 57, No. 4, pp. 125-127.
1922 —— Chemotherapy of antimonial compounds in kala-azar infection. Part I. The toxicity of antimonyl tartrates, &c. Ind. Journ Med. Res., Oct., vol. 10, No. 2, pp. 402-522.
1922 —— Dermal leishmanoid (Editorial). Ind. Journ. Med., March, pp. 31-32.
1923 —— Dermal leishmanoid. Journ. Trop. Med. and Hyg., June 1, vol. 26, No. 11, pp. 182-183.
1923 —— Part II. Dermal leishmanoid. Ind. Journ. Med. Res., April, vol. 10, No. 4, pp. 948-952.
1923 —— Part III. Further observations on the toxicity of antimonyl compounds. Delayed antimony poisoning. Ind. Journ. Med. Res., July, vol. 11, No. 1, pp. 196-213.
1923 —— Part IV. Further observations on the therapeutic value of urea stibamine. Ind. Journ. Med. Res., Oct., vol. 11, No. 2, pp. 393-404.
1923 —— Part V. Amino-antimonyl tartrates. Ind. Journ. Med. Res., Oct., vol. 11, No. 2, pp. 405-410.
1923 —— Part VI. Cumulative and tolerance experiments with tartar emetic. Ind. Journ. Med. Res., Oct., vol. 11, No. 2, pp. 411-415.
1923 —— Urea stibamine in kala-azar. Cal. Med. Journ., Nov., vol. 18, No. 5, pp. 481-490.
1924 —— Chemotherapy of antimonial compounds in kala-azar infection. Part IX. Treatment of kala-azar resistant to antimonial tartrates with urea stibamine. The therapeutic value of stibamine in kala-azar. Ind. Journ. Med. Res., April, vol. 11, No. 4, pp. 1205-1217.
1924 —— Chemotherapy of antimonial compounds in kala-azar infection. Part XI. The value of urea stibamine in the treatment of early kala-azar. Ind. Journ. Med. Res., October, vol. 12, No. 2, pp. 397-402.

1925 BRAHMACHARI, U. N. Chemotherapy of antimonial compounds in kala-azar infection. Part XVII. Further details of the preparation of urea stibamine. Ind. Journ. Med. Res., July, vol. 13, pp. 111-112.
1925 —— Kala-azar, its treatment. Butterworth and Co., 2nd Edition (enlarged).
1926 —— Kala-azar (Innere oder viszerale Leishmaniasis). Handbuch der Tropenkrankheiten, herausgegeben von Prof. Dr. Carl Mense, Kassel, 3 auflage, band 4, Ambrosius Barth, Leipzig.
1924 —— and DAS, J. Chemotherapy of antimonial compounds in kala-azar infection. Part XII. Some observations on the constitution of urea stibamine and stibamine. Ind. Journ. Med. Res., October, vol. 12, No. 2, pp. 423-426.
1925 —— —— Chemotherapy of antimonial compounds in kala-azar infection. Part XV. Further observations on certain derivatives of p-amino-phenyl-stibinic acid. Ind. Journ. Med. Res., vol. 13, pp. 17-19.
1926 —— —— Chemotherapy of antimonial compounds in kala-azar infection. Further observations on certain derivatives of p-amino-phenyl-stibinic acid continued. Part XVIII. Ind. Journ. Med. Res., vol. 13, pp. 693-694.
1923 —— —— and SEN, P. Part VII. The estimation of small quantities of antimony in the presence of organic matter. Ind. Journ. Med. Res., Oct., vol. 11, No. 2, pp. 417-419.
1926 —— —— and BANERJEE, S. C. Chemotherapy of antimonial compounds in kala azar infection. Part I. (New series.) The therapeutic value of N-phenyl-glycine amide-p stibinate of sodium. A preliminary note. Ind. Journ. Med., June, pp. 61-63, and Cal. Med. Journ., pp. 507-510.
1927 —— and DUTT, A. M. Chemotherapy of antimonial compounds in kala-azar infection. Part 3. (New series.) Dermal leishmanoid with positive flagellate culture from the peripheral blood. Ind. Journ. Med., February, pp. 4 6, and Cal. Med. Journ., February, pp. 401-404.
1927 —— —— Dermal leishmanoid with positive flagellate culture from the peripheral blood. Journ. Trop. Med. and Hyg., June 15, vol. 30, No. 12, pp. 158-150.
1927 —— —— Chemotherapy of antimonial compounds in kala-azar infection. Dermal leishmanoid with positive flagellate culture from the peripheral blood. Part 3 (new series), Cal. Med. Journ., vol. 21, pp. 401-404.
1920 —— and DUTT, M. M. A preliminary note on the globulin, albumin and cholesterin contents of the blood in kala-azar. Ind. Journ. Med., vol. 4, No. 1, pp. 281 282.
1924 —— and MAITY, B. B. Chemotherapy of antimonial compounds in kala-azar infection. Part XIV. Observations on a series of cases of kala-azar treated with urea stibamine during a course of 32 hours to 7 days. Ind. Journ. Med. Res., 1924 25, April, vol. 12, No. 4, pp. 735 740.
1925 —— —— Chemotherapy of antimonial compounds in kala-azar infection. Part XVI. Observations on blood-culture of kala-azar patients on N N N medium during 1922-24. I. Comparative value of peripheral blood culture, spleen blood-culture and spleen puncture in the diagnosis of kala-azar. II. The period at which sterilization of the peripheral blood takes place during treatment with urea stibamine. Ind. Journ. Med. Res., vol. 13, pp. 21-24.
1926 —— —— Chemotherapy of antimonial compounds in kala azar infection. Part 2. (New series.) The toxicity and therapeutic value of old samples of urea stibamine. Ind. Journ. Med., vol. 7, September, pp. 130-132, and Cal. Med. Journ., August, pp. 49 51.
1923 —— and SEN, P. The globulin opacity test in kala azar. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 295-296.
1924 —— —— Chemotherapy of antimonial compounds in kala azar infection. Part X, Further observations on quantitative studies in excretion of antimony. The influence of the basic radicle and of repeated injections of an antimonyl tartrate upon the excretion of antimony. Ind. Journ. Med. Res., vol. 12, No. 1, July, pp. 113-126.
1924 —— CHOUDHURY, S. C., DAS, J., and SEN, P. Chemotherapy of antimonial compounds in kala-azar infection. Part VIII. Quantitative studies in the excretion of antimony (tartar emetic and urea stibamine). Ind. Journ. Med. Res., January, vol. 11, No. 3, pp. 820-838.
1921 BRAVO, J., and FRIAS. Another Madrid child with kala-azar. Arch. Españoles de Pediatria, Madrid, July, vol. 5, No. 7, p. 403.
1917 BRIAND. Un cas de kala-azar. Bull. Soc. Méd. Chirurg., Indochine, Dec., vol. 8, No. 2, pp. 54-56.
1914 BRIGHENTI, G. Il kala-azar infantile. Rivista Crit. di Clin. Med., June 13, vol. 15, No. 24, pp. 381-383.
1912 BRITISH MEDICAL JOURNAL. Transmission of kala-azar by bugs. March 9, No. 2671, p. 567.

1904 —— Vol. 1, pp. 861-862. (Leishman-Donovan bodies in kala-azar: Dr. Bentley's observation.)
1904 —— Vol. 1, p. 911. The Leishman-Donovan parasite (Editorial).
1904 —— Vol. 2, pp. 642-658. Discussion on the Leishman-Donovan body.
1917 BRODIE, F., and YORKE, W. A case of kala-azar in the Mediterranean Expeditionary Force. Journ. Roy. Army Med. Corps, January, vol. 28, No. 1, pp. 91-97.
1908 BROOKE, G. E. Tropical medicine, hygiene and parasitology. Charles Griffin and Co., Ltd., Kala-azar, pp. 237-246.
1927 BROOKE, ROGER. Kala-azar, report of a case of. Amer. Journ. Trop. Med., January, vol. 7, No. 1, pp. 27-38.
1898 BROWN, E. H. A report on kala-dukh, a form of fever met with in the north-east portion of the district of Purnea. Ind. Med. Gazette, vol. 33, pp. 324-330.
1904 BRUCE, DAVID. Discussion on the Leishman-Donovan body. Brit. Med. Journ., vol. 2, p. 658.
1906 BUCHANAN, R. M. Demonstration of specimens from a case of kala-azar in a European. Glasgow Pathological and Clinical Society. Lancet, vol. 1, No. 4305, pp. 603 604.
1912 BURGESS, J. H. Leishmania and mosquitoes (correspondence). The Lancet, vol. 182, No. 4611, p. 123.
1925 BURNET, E., and MASSELOT, F. Case of kala-azar in the adult in Tunis. Bull. et Mém. Soc. Méd. d. Hôp. de Paris, May 13, vol. 40, pp. 307-309.
1925 —— —— Deuxième cas de kala-azár chez l'adult en Tunisie. Bull. Soc. Path. Exot., vol. 18, pp. 297-300. Un cas tunisien de kala-azár chez l'adults. Bull. et Mém. Soc. Méd. d. Hôp. de Paris, vol. 49, p. 3.
1925 —— —— CALAMIDA, F., and ORTONA, C. Trois cas de kala-azár chez l'adults en Tunisie. Arch. Inst. Pasteur de Tunis, vol. 14, pp. 331-330.
1922 DE BUEN, SADI. Un caso de kala-azar en la Provincia de Cáceres. Arch. de Cardiologia y Hematologia, Madrid, June, vol. 3, No. 6, pp. 205-208.
1922 —— Algunas consideraciones sobre el kala-azar en España. Rev. de Hyg. y d. Tuberculosis. Oct. 31, Year 15, No. 173, pp. 253-259.
1923 —— Apunte sobre la naturaleza de las celulas parasitiferas de los bazos leishmaniosicos. Reprinted from Arch. de Cardiologia y Hematologia. Vol. 4, No. 1, p. 2.
1926 —— Le kala-azar infantile et les autres leishmanioses en Espagne. Bull. Office Internat. d'Hyg. Publique, vol. 18, pp. 268-270.
—— El kala azar infantil. Instrucciones para su diagnostico y tratamiento. 2a edicion. Ministerio de la Gobernacion Publicaciones de la Direction-General de Sanidad, Madrid, pp. 8, with 5 figs.
1905 BUSHNELL, F. G. *Spirochæta pallida* and Leishman-Donovan bodies. The Lancet, vol. 2, p. 1728.

1902 CADUCÉE, LE. Vol. 2, p. 65. Le Ponos.
1912 CALAMIDA, F. Vingt-Sixième observation Tunisienne de kala-azar. Archives de l'Institut Pasteur de Tunis, vol. 2, p. 61.
1912 —— Vingt-neuvième observation Tunisienne de kala-azar. Archives de l'Institut Pasteur de Tunis, vol. 2, pp. 63-64.
1923 CALCUTTA MEDICAL JOURNAL. January (kala-azar number.)
1922 CAMPANA Y CASSI, T. Le kala-azar infantile dans la Province de Tarragone. Rev. Méd. et Hyg. Trop., May-June, vol. 14, No. 3, pp. 71-72.
1924 CAMPOS, J. J. Leishmaniosis in Goa. (Conferencia feits na A.M.P.I.P.). Bol. Ger. Med. e Farmacia, Castona, November, pp. 430-445.
1910 CANNATA, S. Contributo alla conoscenza dell' anemia splenica infantile da parassiti di Leishman. Riforma Medica, June 6, vol. 26, No. 23, pp. 624-626.
1910 —— Ricerche ematologiche nell' anemia splenica infantile da parassiti di Leishman. La Pediatria, 2 series, vol. 8, pp. 321-338.
1910 —— Seconda serie di ricerche ematologiche nell' anemia da leishmania. La Pediatria, 2 series, vol. 8, pp. 718 724.
1911 —— Terza serie di ricerche ematologiche nell' anemia da leishmania. La Pediatria, 2 series, vol. 9, pp. 481-484.
1911 —— Qualche considerazione a proposito di un nuovo caso di anemia da leishmania. xxiii, osservato a Palermo. Riforma Medica, Palermo, March 6, vol. 27, No. 10, pp. 263-264.
1911 —— Il potere fagocitario del sangue nell' anemia da leishmania. Gazzetta Internazionale di Medicina, vol. 14, p. 5.
1912 —— Sul potere agglutinate del siero di sangue nell' anemia da leishmania rispetto ad alcuni germi patogeni. Pathologica, August 15, vol. 4, No. 91, p. 482.

1912 CANNATA, S. Quarta serie di ricerche ematologiche nell' anemia da leishmania. La Pediatria, No. 8.
1913 —— *Sul reperto del parassita di leishman nel sangue periferico.* Malaria e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 303-306.
1913 —— Inclusioni leucocitarie nella leishmaniosi infantile. Pathologica, July 15, vol. 5, No. 113, p. 420.
1913 —— Le piastrine del sangue nella leishmaniosi infantile. Pediatria, Sept. 30, vol. 21, No. 9, pp. 645-648.
1913 —— Reperto del parassita di Leishman nel sangue periferico. Nota Preventiva. Pathologica, June 15, vol. 5, No. 111, pp. 351-352.
1913 —— Sul reperto del parassita di Leishman nel sangue periferico. Riforma Med., August 2, vol. 29, No. 31, pp. 844-846.
1914 —— Ulteriori ricerche sulla presenza del parassita di Leishman nel sangue periferico di bambini affetti da leishmaniosi. Pediatria, January, vol. 22, No. 1, pp. 27-32.
1921 —— Leishmaniosi interna. Relazioni al VI Congresso Medico Siciliano. Riv. San Siciliana, No. 9, year 27, vol. 2, Nos. 3-4, pp. 651-652.
1925 —— Ulteriore contributo alls terapia della leishmaniosi infantile. Pediat., vol. 33, pp. 1309-1311.
1925 —— Contributo alla terapia della leishmaniosi infantile. Pediatria, May 1, vol. 33, No. 9, pp. 440-452.
1925 —— Treatment of infantile leishmaniasis. Pediatria, December 15, vol. 33, No. 24, pp. 1309-1311.
1914 —— and CARONIA, G. Cultura dei parassiti di Leishman dal sangue periferico nella leishmaniosi infantile. Pathologica, August 1, vol. 6, No. 138, pp. 306-308.
1926 DE CAPUA, F. La diffusions della leishmaniosi infantile. Observazione sulla casistica di un decennio. Pediatria, May 1, vol. 34, pp. 440-466.
1924 DE CAPITE, A. Sull gelatin del siero di sangue nella leishmaniosi infantile. Pediatria, April 1, vol. 32, No. 7, pp. 408-413.
1911 CARDAMATIS, J. P. Des piroplasmiases et leishmaniases. Central f. Bakt. 1 Abt. Orig., October 21, vol. 9, part 6, pp. 511-523.
1912 —— Leishmaniose du chien en Grèce. Bulletin de la Soc. Path. Exot., Séance of February 14, vol. 5, No. 2, pp. 88-89.
1912 —— Le kala-azar est en Grèce une maladie à cas sporadiques. Mégalosplénies de cause inconnue. Bull. Soc. Path. Exot., July, vol. 5, No. 7, pp. 480-491.
1911 —— and MELISSIDIS, APOLLODORE. Du rôle probable de la mouche domestique dans la transmission des leishmania. Bulletin de la Société de Pathologie Exotique, July 12, vol. 4, No. 7, pp. 459-461; Nos. 15-16, pp. 258-262.
1880 CARDARELLI. Della pseudoleucemia splenica dei bambini. (Congresso Medico di Genova.)
1890 —— Nosografia della pseudoleucemia splenica (infettiva) dei bambini. Bollettino della reale Accademia Medico-chirurgica di Napoli, vol. 2, pp. 17-44.
1921 CARNOT, P., and LIBERT, E. Un cas de kala-azar, d'origine Macédonienne, survenu chez un adulte et observé à Paris. Bull. et Mém. Soc. Méd. Hôpit. de Paris, July 14, No. 24, pp. 1030-1046.
1905 CARONIA and DI GIORGIA, G. Sulla leishmaniosi spontanea nei cani di Palermo. Pathologica, 1914, April 15, vol. 6, No. 131, pp. 208-209.
1912 —— Tentative d'immunizzazione attiva per la leishmaniosi umana nel bambino sano. Pathologica, Dec. 1, vol. 4, No. 98, pp. 724-725.
1913 —— Agglutinine e precipitie specifiche nella leishmaniosi infantile. Pediatria, Sept. 30, vol. 21, No. 9, pp. 641-644.
1913 —— Curve termiche nella leishmaniosi infantile. Pediatria, July 31, vol. 21, No. 7, 2 serie, vol. 11, pp. 481-496.
1913 —— Sulla guaribilità dell' anemia da leishmania (a proposito di 8 casi di guarigione osservati a Palermo). Malaria e Malat. d. Paesi Caldi, March, vol. 4, No. 2, pp. 90-96.
1913 —— Weiterer Beitrag zur Leishmania Anämie. Arch. f. Kinderheilkunde, Feb. 20, vol. 59, Nos. 5-6, pp. 321-347.
1913 —— L'anafilassi nella leishmaniosi infantile. Pathologica, July 15, vol. 5, No. 113, pp. 420-423.
1913 —— Spezifische Agglutinine und Präzipitine bei der infantilen Leishmaniosis. Zeitschr. f. Immunitätsforsch. u. Experiment. Therapie, 1 Teil. Orig., Nov. 24, vol. 20, Nos. 1-2, pp. 174-177.
1913 —— Sul potere complementare del siero di sangue nella leishmaniosi infantile. Pediatri, August 31, vol. 21, No. 8 (ser. 2, vol. 11), pp. 583-587; Mal. e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 309-313.
1913 —— Über die Heilbarkeit der Leishmania-Anämie. Zeitschr. f. Kinderheilk. Orig., Sept. 13, vol. 8, No. 6, pp. 452-460.

1914 —— Fieberkurven bei der kindlichen Leishmaniosis. Deutsch. Arch. f. klinische Medizin, January 31, vol. 113, Nos. 3-4, pp. 354-371.
1916 —— L'impiego di nuovi preparati di antimonio per via intramusculare nella cura della leishmaniosi infantile. Pediatria, Feb., vol. 24, No. 2, pp. 65-81.
1922 —— De la ponction de la rate et de la moelle osseuse. Bull. Soc. Path. Exot., Oct. 11, vol. 15, No. 8, pp. 722-729.
1910 CARISTO, F. Contributo allo studio del kala-azar in Calabria. Studi intorno ad alcune malattie tropicali in Sicilia e Calabria. Tipografia Labicana, vol. 1.
1926 CARRILLO, F. Nasapharyngeal form of kala azar. Rev. Méd. del Rosario, vol. 16, pp. 458-464.
1922 CARROSCO, M. C. Revista sintetica sobre as leishmanioses viscerais. Reprinted from Med. Contemp., p. 54.
1912 CARYOPHYLLIS, G., and SOTIRIADES, D. Zur Behandlung und Heilung des Kala-Azar mit Salvarsan. Deut. med. Wochenschr., August 15, vol. 38, No. 33, p. 1554.
1911 —— —— Zur Kasuistik des Kala-azar und seiner Behandlung mit Salvarsan. Deutsch. med. Wochenschrift, October 12, vol. 37, No. 41, pp. 1896-1898.
1912 —— —— Kala azar. Archives de Médecine, March 1-10, 8me année, Nos. 7-8, pp. 68-71.
1903 CASTELLANI, ALDO. "Leishman-Donovan" bodies in Ceylon. Archiv. für Schiffs- und Tropenhygiene, vol. 8, pp. 464-466.
1904 —— Leishmania donovani in Ceylon. Brit. Med. Journ., vol. 2, pp. 656-557; Journ. Trop. Med. and Hyg., August 15, p. 262.
1915 —— Breve nota sulla cura di un caso di kala-azar. Pediatria, April, vol. 23, No. 4, pp. 241-243.
1915 —— Treatment of "Flagellate diarrhœa" and of kala-azar. Brit. Med. Journ., Nov. 27, pp. 779-780.
1915 —— Ditto. Journ. Trop. Med. and Hyg., May 15, vol. 18, No. 10, pp. 112-113.
1919 —— and CHALMERS, ALBERT J. Manual of Tropical Medicine, 3rd edition.
1922 CASTORINA, G. Sul contenuto in colesterina del siero di sangue nella leishmaniosi infantile. Pediatria, vol. 30, Nov. 15, No. 22, pp. 1076-1080.
1922 —— Cronaca della leishmaniosi infantile a Messina e dintorni. Pediatria, Dec. 15, vol. 30, No. 24, pp. 1173-1178.
1922 —— Un mode d'inoculation de la leishmaniosi cutanée. Rev. Méd. et Hyg. Trop., July-August, vol. 15, No. 4, pp. 135-138.
1923 —— La reazione di Brahmachari nella leishmaniosi infantile. Pediatria, Nov. 1, vol. 31, No. 21, pp. 1172-1179.
1923 —— Lo schema di Arneth nella leishmaniosi infantile prima e durante la terapia antimoniale. Pediatria, Dec. 1, vol. 31, No. 23, pp. 1270-1277.
1924 —— Un caso di leishmaniosi infantile a decorso molto protratto. Pediatria, Feb. 1, vol. 32, No. 3, pp. 174-176.
1905 CATHOIRE. Observation d'un cas de piroplasmose généralisée en Tunisie. Archives Générales de Médecine, vol. 1, pp. 1426-1427.
1925 CESARI, E. La leishmaniose canine. Rev. Générale Méd. Vét., vol. 34, pp. 612-632.
1926 —— La leishmaniose canine en France. Recueil Méd. Vét., vol. 101, pp. 177-179.
1927 CHADWICK, C. R., and MCHATTIE, C. Notes on cutaneous leishmaniasis of dogs in Iraq. Trans. Soc. Trop. Med. and Hyg., vol. 20, pp. 422-432.
1925 CHATTERJEE, A. C. The race, class, sex and age incidence of kala-azar. Ind. Med. Rec., vol. 45, pp. 129-131.
1926 —— Experience in the treatment of kala-azar with urea stibamine. Ind. Med. Rec., vol. 46, pp. 30-44.
1926 —— Infectiousness of kala-azar and its control. Ind. Med. Rec., vol. 46, pp. 1-14.
1905 CHATTERJEE, GOPAL CHANDRA. The cultivation of trypanosoma out of the Leishman-Donovan body upon the method of Capt. L. Rogers. Lancet, January 7, vol. 1, p. 16.
1909 —— Etiology of double quotidian fever with some notes on the early stage of Leishman-Donovan infection. Paper read before the Medical Section of the Asiatic Society, April, Ind. Med. Gazette, Sept., vol. 44, pp. 334-336.
1923 —— Control of kala-azar in Bengal. Ind. Med. Record, August, pp. 205-208.
1923 —— Kala-azar in Bengal. (Correspondence.) Ind. Med. Gaz., November, vol. 58, No. 11, pp. 553-554.
1927 —— and BOSE, S. C. Control of kala-azar by mass treatment with antimony. Ind. Med. Record, September, pp. 269-271.
1923 CHATTERJEE, H. An unusual complication in kala-azar. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 313-314.

1925 CHATTERJEE, K. N. A case of kala azar with transposition of viscera. Ind. Med. Gazette, vol. 60, p. 326.
1919 CHATTON, EDOUARD. Sur la culture pure d'un leptomonas de la puce du chien et sur un caractère de ses formes culturales qui les distinguent de celles du kala-azar de souches humaine et canine. Bull. Soc. Path. Exot., June 11, vol. 12, No. 6, pp. 313-316.
1905 CHILDE, L. F. The first case of Leishman-Donovan disease occurring in a European in Bombay. Ind. Med. Gazette, vol. 11, pp. 449-477.
1926 CHOPRA, R. N. A preliminary note on the pharmacological action of certain organic antimony derivations. Ind. Med. Gazette, vol. 61, No. 4, pp. 162-166.
1927 —— An experimental investigation into the action of organic compounds of antimony. Ind. Journ. Med. Res., July, vol. 15, No. 1, pp. 41-48.
1927 —— GUPTA, J. C., and BASU, N. K. Further observations on the serum test for kala-azar with organic antimony compounds. A simple blood-test for kala-azar. Ind. Med. Gazette, August, vol. 62, No. 8, pp. 434-437.
1927 —— —— —— Antimony test for diagnosis of kala-azar. Ind. Med. Gazette, December, vol. 62, No. 12, pp. 688-691.
1927 CHOPRA, R. N., in collaboration with GUPTA, J. C., and DAVID, J. C. A preliminary note on the action of antimony compounds on the blood serum. A new serum test for kala-azar. Ind. Med. Gaz., June, pp. 325-327.
1911 CHRISTOMANOS, ANT. A. Kala-azar-fälle in Griechenland. Deutsche med. Wochenschr., April 6, vol. 37, No. 14, pp. 641-644.
1911 —— Über den therapeutischen Wert des Salvarsans bei Kala-azar. Deut. med. Wochensch., Sept. 14, vol. 37, No. 37, pp. 1705-1706.
1904 CHRISTOPHERS, S. R. A preliminary report on a parasite found in persons suffering from enlargement of the spleen in India. Scientific memoirs by Officers of the Medical and Sanitary Departments of the Government of India. New series, No. 8, pp. 1-17.
1904 —— On a parasite found in persons suffering from enlargement of the spleen in India. Second Report. Scientific memoirs by Officers of the Medical and Sanitary Departments of the Government of India. New series, No. 11, pp. 1-21.
1904 —— Tropical splenomegaly and oriental sore. Brit. Med. Journ., Sept. 17, vol. 2, pp. 655-656; Journ. Trop. Med. and Hyg., August 15, pp. 261-262.
1905 —— On a parasite found in persons suffering from enlargement of the spleen in India. Third Report. Scientific memoirs by Officers of the Medical and Sanitary Departments of the Government of India. New series, No. 15, pp. 1-14.
1911 —— An investigation into the prevalence of kala-azar in a part of Upper Assam. Paludism. July, No. 3, pp. 72-86.
1926 —— Recherches sur le kala azar dans l'Inde Britannique. Bull. Office Internat. Hyg. Publique, vol. 18, pp. 271-272.
1926 —— The result of intra-peritoneal inoculation of mice with cultures of *Leishmania donovani*. February, Indian Medical Research Memoir, pp. 77-87.
1926 —— The result of feeding mice with cultures of *Herpetomonas donovani*. February, Indian Medical Research Memoir, pp. 89-103.
1926 —— Development of parasite of Indian kala-azar in the sandfly *Phlebotomus argentipes*: controls. February, Indian Medical Research Memoir, pp. 135-130.
1926 —— Development of the parasite of Indian kala-azar in the sandfly *Phlebotomus argentipes*: re-fed flies and further results of the feeding of sandflies on kala-azar cases. February, Indian Medical Research Memoir, pp. 141-145.
1926 —— The feeding of larvæ of *Phlebotomus argentipes*, Annandale and Brunetti, on cultures of *Leishmania donovani*, February, Indian Medical Research Memoir, pp. 155-156.
1926 —— Technique employed in breeding *Phlebotomus argentipes* in Assam. February, Indian Medical Research Memoir, pp. 173-175.
The anatomy of the sandfly *Phlebotomus argentipes*, Annandale and Brunetti (Diptera). 1. The head and mouth parts of the imago. February, Indian Medical Research Memoir, pp. 177-204.
1926 —— Introduction. Ind. Journ. Med. Res. Memoir No. 4, pp. 3-18.
1925 —— SHORTT, H. E., and BARRAUD, P. J. Culicoides and kala-azar in Assam. Ind. Journ. Med. Res., 1925-1926, July, vol. 13, pp. 175-176.
1925 —— —— —— Our present knowledge regarding the transmission of kala-azar in nature. Proc. Assam Branch Brit. Med. Assoc. Ann. Meeting, Jorhat, March 2, pp. 22-28.
1925 —— —— —— Temperature in relation to culture of *Herpetomonas donovani* on N N N medium. Ind. Journ. Med. Res., July, vol. 13, No. 1, pp. 167-171.
1925 —— —— —— Further observations on the feeding of sandflies, *Phlebotomus argentipes*, on cases of Indian kala-azar. Ind. Journ. Med. Res., vol. 13, pp. 159-165.

1925 —— —— —— Effect of salt solutions of different concentrations on parasite of Indian kala-azar. Ind. Journ. Med. Res., vol. 13, pp. 177-182.
1925 —— —— —— The development of the parasite of Indian kala-azar in the sand-fly, *Phlebotomus argentipes*. Ind. Journ. Med. Res., January, vol. 12, No. 3, pp. 605-607.
1926 —— —— —— The morphology and life-cycle of the parasite of Indian kala-azar in culture. February, Indian Medical Research Memoir, pp. 19-53.
1917 CHRISTOPHERSON, J. B. Notes on a case of espundia (naso-oral leishmaniasis) and three cases of kala-azar in the Sudan treated by the intravenous injection of antimonium tartaratum. Journ. Trop .Med. and Hyg., October 15, vol. 20, No. 20, pp. 229-236.
1922 —— The "blue bodies" in leishmaniasis. Proc. Roy. Soc. Med. (Sect. Trop. Disease and Parasitology), April, vol. 15, No. 6, pp. 21-24.
1913 CIMINATA, A. Sul reperto parassitario nelle anemie da leishmania. Rivista Ospedaliera, Feb. 28, vol. 3, No. 4, pp. 156-159.
1882 CLARKE, J. J. Annual Report of the Sanitary Commissioner with the Government of Assam. Shillong.
1911 COCHRAN, S. Kala-azar infantum in Hwaiyuan. (Correspondence.) China Med. Journ., July, No. 4, pp. 272-273.
1911 —— Leishmaniasis in China. (Correspondence.) Journ. Amer. Med. Assoc., Nov. 11, vol. 57, No. 20, pp. 1630-1631.
1912 —— The superficial lymph nodes as a source of leishmania for diagnosis in kala-azar. Journ. Trop. Med. and Hyg., Jan. 1, vol. 15, No. 1, pp. 9-10.
1913 —— The superficial lymph nodes as a source of leishmania for the diagnosis of kala-azar with some observations of kala-azar in China. Journ. Lond. School Trop. Med., November, vol. 2, part 3, pp. 179-195.
1915 —— Splenectomy in kala-azar. China Med. Journ., Sept., vol. 29, No. 5, pp. 301-307.
1922 COMBIESCO, D. Researches sur la gelification du serum par l'aldehyde formique chez des animaux an état d'anaphylaxie. C. R. Soc. Biol., July 8, vol. 87, pp. 416-417.
1914 CONOR, A., and CALÒ, E. Le troisième cas de kala-azar d'origine Algérienne. Bull. Soc. Path. Exot., Jan., vol. 7, No. 1, pp. 42-43.
1912 CONSEIL, E. Contribution à la vingt-cinquième observation Tunisienne de kala-azar. Traitement par l'hectine. Archives de l'Institut Pasteur de Tunis, vol. 2, p. 65.
1916 CORNWALL, J. W., and LA FRENAIS, H. M. A contribution to the study of kala-azar. Ind. Journ. Med. Res. (I), April, vol. 3, No. 4, pp. 680-724.
1916 —— —— A contribution to the study of kala-azar (II). Ind. Journ. Med. Res., July, vol. 4, No. 1, pp. 105-119.
1922 —— —— A contribution to the study of kala-azar (V). Ind. Journ. Med. Res., January, vol. 9, No. 3, pp. 533-544.
1923 —— —— Note on the resistant forms of *Leishman donovani* found in N N N cultures. Ind. Journ. Med. Res., vol. 11, pp. 587-589.
1917 —— and MENON, T. K. A contribution to the study of kala-azar (III). Ind. Journ. Med. Res., April, vol. 4, No. 4, pp. 672-687.
1918 —— —— Ditto. (IV). Ind. Journ. Med. Res., April, vol. 5, No. 4, pp. 541-547.
1910 CORTESI, A., LEVY, E., and ORTONA, C. Recherches sur le kala-azar infantile de Tunisie entreprises a l'Institut Pasteur de Tunis.
I. Dix-septième observation Tunisienne de kala-azar infantile; traitement par les injections intraveineuses de sublime; Insuccès. par A. Cortesie and E. Levy.
II. Contribution à la quinzième observation tunisienne de kala-azar infantile; traitement par l'emétique d'aniline; Insuccès. par E. Levy and C. Ortona. Archives de l'Institut Pasteur de Tunis, le fasc, pp. 13-18.
1910 —— —— TRIOLO, G., CONOR, A., NICOLLE, C., COMTE, C., and MANCEAUX, L. Recherches sur le kala-azar infantile de Tunisie entreprises à l'Institut Pasteur de Tunis.
I. Dix-huitième observation tunisienne de kala-azar infantile traitement par l'électro-mercurol. A. Cortesi and E. Levy.
II. Dix-neuvième observation tunisienne de kala-azar infantile; traitement par l'électro-mercurol; noma. A. Cortesi and E. Levy.
III. Vingtième observation tunisienne de kala-azar infantile. G. Triolo.
IV. Essai de traitement du kala-azar expérimental du chien par le mercure colloidal. Resultat negatif. A. Conor.
V. Kala-azar expérimental du chien. Quelques observations nouvelles et quelques faits nouveaux. Immunité par première atteinte après guérison complète. C. Nicolle and C. Comte.

VI. Inoculation sous-cutanée du virus du kala-azar au chien et au singe. C. Nicolle and L. Manceaux.
VII. Technique de la ponction du foie chez le chien. C. Nicolle and A. Conor.
VIII. Etat actuel de la question du kala-azar infantile. C. Nicolle. Archives de l'Institut Pasteur de Tunis, 3e part, pp. 99-114.

1912 —— Trentième observation tunisienne de kala-azar. Archives de l'Institut Pasteur de Tunis, vol. 2, pp. 62-64.

1912 —— Vingt-huitième observation tunisienne de kala-azar. Arch. de l'institut Pasteur de Tunis, part 2, pp. 62-63.

1916 COSTA, P., Jr. Nota preliminar sobre lesoes oculares da leishmaniosi. Gaz. Med., Bahia, June, vol. 47, No. 12, pp. 496-498.

1913 CRAGG, F. W. An investigation into kala-azar. Proc. 3rd Meet. Genl. Mal. Com. (Madras), 1912. Pp. 39-42.

1911 CRAIG, C. F. Kala-azar, infantile splenic anæmia and oriental sore (leishmaniosis). *Modern Treatment*, Hare. Philadelphia and New York. Vol. 2, pp. 61-66.

1913 CRESPIN. Leishmaniose et paludisme chronique infantile. Caducée. April 5, vol. 13, No. 7, p. 89.

1910 DI CRISTINA, G. Su alcuni nuovi casi di anemia da leishmania osservati in Palermo. La Pediatria, series 2, vol. 8, No. 11, pp. 832-843.

1911 —— La courbe fébrile le plus fréquemment observée en Sicile, dans l'anémie splénique dûe au parasite de Leishman. Revue d'Hygiéne et de Médecine Infantiles, vol. 10, pp. 46-59.

1911 —— Sulle culture dei parassiti di Leishman nel terreno di Novy Neal e Nicolle. Pathologica, May 1, vol. 3, No. 60, p. 206.

1912 —— I corpi Leishman nell' organismo infetto vanno incontro a processi litici (Nota Preventiva). Pathologica, Nov. 1, vol. 4, No. 96, pp. 643-645.

1912 —— Ancora sulla deviazione del complemento nella anemia da leishmania. Pathologica, July 15, vol. 4, No. 89, pp. 409-410.

1913 —— Serologische Untersuchungen bei der infantilen leishmaniosis. Ztschr. f. Kinderheilk, Nov. 15, vol. 9, No. 2, pp. 128-140.

1914 —— Complicanze dell' Apparato digerente nella leishmaniosi interna. Lavori d. Soc. Italiana di Patalogia Esotica, pp. 80-92.

1915 —— Über die Behandlung der inneren Leishmaniosis. Deutsche med. Wochenschr., April 1, vol. 41, No. 14, pp. 396-397.

1921 —— Terapia della leishmaniasi interna. Relazioni al VI Congresso Medico Siciliana, No. 9. (Summarized in Ann. di Med. Nav. e Colon., 1921, Sept.-Oct., year 27, vol. 2, Nos. 3-4, p. 652.)

1910 —— and CANNATA, S. Sui caratteri morfologici e culturali del parassita dell' anemia splenica infantile *Leishmania infantum*. Gazzetta Degli Ospedali e Delle Cliniche, Milano, vol. 31, pp. 505-506.

1910 —— —— Über die morphologischen und kulturellen Eigenschaften des Parasiten der infantilen Milzanämie (*Leishmania infantum*). Centralblatt für Bakteriologie, Jena, Part I Orig., vol. 55, No. 6, pp. 494-496.

1910 —— —— Ricerche anatomo patologiche in un caso di anemi splenica infantile da parassiti di Leishman. La Clinica Medica Italiana, vol. 40, pp. 493-503. Milan: Casa ed. Dr. F. Vallardi.

1910 —— —— Sul ricambio respiratorio nell' anemia splenica infantile da parassiti di Leishman. Rivista di Clinica Pediatrica, vol. 7, No. 6, p. 472 sqq.

1911 —— —— Infezione sperimentale dei cani con la leishmania umana. Pathologica, May 15, vol. 3, No. 61, pp. 233-234.

1911 —— —— Contributo allo studio dell' anatomia patologica dell' anemia da leishmania. La Pediatria, series 2, vol. 9, No. 3, pp. 200-211.

1912 —— and CARONIA, G. Primi tentativi di vaccinazione graduale nell' anemia da leishmania von culture morte (Nota Preventiva). Pathologica, Sept. 1, vol. 4, No. 92, pp. 519-522.

1912 —— —— Sulla presenza di ambocettori specifici in bambini affetti da anemia da leishmania guariti spontaneamente. Pathologica, Sept. 1, vol. 4, No. 92, pp. 534-535.

1913 —— —— Ricerche serologiche nella leishmaniosi infantile. Pediatria, Nov. 30, vol. 21, No. 11, pp. 801-817.

1915 —— —— Über die Therapie der inneren Leishmaniosis. Deutsch. Arch f. klin. Med., vol. 117, pp. 263-277.

1915 —— —— Sulla terapia della leishmaniosi interna. Pathologica, February 15, vol. 7, No. 151, pp. 82-83, and Bull. Soc. Path. Exot., Feb., vol. 8, No. 2, pp. 63-66.

1915 —— —— Sulla terapia della leishmaniosi interna. Pediatria, Feb., vol. 23, No. 2, pp. 81-92.
1916 —— —— Terapia della leishmaniose interna. Mal. et Malat. de Paesi Caldi, July-August, vol. 7, No. 4, pp. 245-253.
1910. CRITIEN, A. Kala-azar infantile à Malte. Note préliminaire. Archives de l'Institut Pasteur de Tunis, vol. 2, pp. 49-51.
1911 —— Kala-azar in Malta. Brit. Med. Journ., January 23, No. 2613, p. 198.
1911 —— Infantile leishmaniasis (marda tal biccia) in Malta. Ann. of Trop. Med. and Parasit., April 20, vol. 5, No. 1, pp. 37-56.
1904 CROMBIE, ALEXANDER. Discussion on the Leishman-Donovan body. Brit. Med. Journ., vol. 2, p. 658.
1904 —— Dum-Dum fever? Kala-azar? Non-malarial remittent fever? (Correspondence.) Journ. Roy. Army Med. Corps, vol. 2, p. 509.
1906 CUMMINS, S. LYLE. Kala-azar and its intermediate host: A suggestion. Journ. Roy. Army Med. Corps, vol. 7, pp. 303-304.
1908 —— Kala-azar in the Anglo-Egyptian Sudan. Third Report of the Wellcome Research Laboratories at the Gordon Memorial College, Khartoum, pp. 100-106.
1908 —— Kala azar in the Anglo-Egyptian Sudan. Journ. Roy. Army Med. Corps, vol. 10, pp. 174-176.
1885 CUNNINGHAM, D. D. On the presence of peculiar parasitic organisms in the tissue of a specimen of Delhi boil. Scientific Memoirs by Medical Officers of the Army of India, Part I, 1884, pp. 21-31.
1923 CUNNINGHAM, J., and VARADARANJAN, P. S. Kala-azar in the Madras Presidency outside Madras City. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 308-311.
1923 —— —— Notes on results of treatment of cases of kala-azar admitted to Madras city hospitals between 1913-1922. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 307-308.
1923 —— —— An epidemiological study of 663 cases of kala-azar admitted to the Madras city hospitals between 1913-1922. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 301-306.
1925 —— and PUNDIT, S. R. A new endemic focus of kala azar in Southern India. Ind. Journ. Med. Res., April, vol. 12, No. 4, pp. 743-754.

1907 DALAND, JUDSON. Assam fever or Leishman-Donovan disease. Boston Medical and Surgical Journ., vol. 157, p. 154.
1913 DANIELS, C. W. Tropical Medicine and Hygiene, 2nd edition.
1911 DARLING, S. T. The blood platelets in tropical and other forms of anæmia. Trans. of the Society of Trop. Med. and Hyg., November, vol. 5, No. 1, pp. 46-57. Kala-azar, pp. 53-54.
1923 DAS GUPTA, AMIYA SANKER. Kala-azar at Bogra. Ind. Med. Record, August, pp. 237-238.
1922 —— The diagnosis of kala-azar by peripheral blood-culture (abstract). Ind. Med. Gazette, vol. 57, No. 6, p. 217.
1922 —— A case of antimony-fast leishmaniasis. Ind. Med. Record, August, p. 177.
1927 —— Further observations upon dermal leishmanoid. Ind. Med. Gaz., April, p. 190.
1922 —— A note on some cultural phases of *Leishmania donovani*. Ind. Journ. Med. Res., April, vol. 9, No. 4, pp. 809-813.
1923 —— Antimony-fast kala-azar. Ind. Med. Gazette, January, vol. 58, No. 1, p. 46.
1927 DAS GUPTA, B. M. A note on the parasite of dermal leishmanoid. Ind. Med. Gaz., January, pp. 11-12.
1927 — Further observations upon "Dermal Leishmanoid." Ind. Med. Gaz., vol. 62, p. 190.
1925 DAS SUDHIR, K. Stibosan (von Heyden 471) in private practice. Ind. Med. Gazette, September, vol. 60, pp. 425-426.
1923 D'ASTROS, GIRRAUD, P., and RAYBAUD, J. Quatre cas autochtones de kala-azar infantile observés à Marseilles. Bull. Acad. Méd., July 31, Year 87, vol. 90; third series, No. 31, pp. 114-118.
1913 DAVIS, W. Notes on two cases of kala-azar. Journ. Roy. Army Med Corps, January, vol. 20, No. 1, pp. 87-88.
1909 DAY, H. B., and FERGUSON, A. R. An account of a form of splenomegaly with hepatic cirrhosis, endemic in Egypt. Ann. of Trop. Med. and Parasit., November 1, vol. 3, No. 3, pp. 379-394.
1923 DE, M. N. An unusual complication in an acute case of kala-azar. Ind. Med. Gazette, October, vol. 58, No. 10, pp. 484-485.

1923 DE, M. N. A case of kala-azar with fatal bleeding. Calcutta Med. Journ., March, vol. 17, No. 9, pp. 120-121.

1924 —— A simple method of staining *Leishmania donovani* in tissues. Ind. Med. Gazette, February, vol. 59, No. 2, pp. 82-84.

1927 —— Leishmaniasis as an infection of the reticulo endothelial system. Ind. Journ. Med., vol. 8, part 1, February, pp. 13-17.

1911 DELANOE, P. L'importance de la phagocytose dans l'immunité de la souris à l'égard de quelques flagellés. (Travail de pathologie expérimentale.) Thèse, Montpellier, No. 15. Paris: Imprimerie de la Cour d'Appel, p. 46.

1912 —— Importance de la phagocytose dans l'immunité de la souris à l'égard de quelques flagellés. Ann. de l'Institut Pasteur, March, vol. 26, No. 3, pp. 172-179.

1923 DE SOUZA, SALA-ZAR. La splenectomie dans le kala azar infantile. Revista Med. de Angola (No. Especial 1º Congresso de Med. Trop. da Africa Ocidental), vol. 4, 6a, 7a, 8a Sessoes, August, No. 4, pp. 400-417.

1913 DIONISI, A. Contributo alla anatomia pathologica dell' anemia da leishmania. Mal. e Malat. d. Paesi Caldi, June-July, vol. 4, No. 4, pp. 265-290.

1914 DIVARIS, S. Kala-azar in Cephalonia. *Vide* Trop. Dis. Bull., vol. 4, No. 1, p. 395.

1802 DOBSON, EDWIN. Notes regarding the prevalence of the *Dochmius duodenalis*. Ind. Med. Gazette, December, vol. 27, pp. 354-357; 1893, vol. 28, pp. 1-4, 40-43, 68-72, 202-207.

1894 —— The prevalence of *Dochmius duodenalis* in India. Transactions First Indian Medical Congress, Calcutta, pp. 56-62.

1922 D'OELSNITZ, BALESTRE, P. L., and DANMAS, A. Un nouveau cas de kala-azar infantile en France. Bull. et Mém. Soc. Méd. Hôp. de Paris, April 6, 38th year, 3rd series, No. 12, pp. 550-553.

1925 D'OELSNITZ, DAUMAS, LIOTARD, PUECH. Quatre cas de kala azar d'origine française. Résultats favorables du traitement par les injection intraveineuses d'émétique de soude. Bull. et Mém. Soc. Méd. Hôpit. de Paris, Feb. 26, year 41, 3rd ser., vol. 50, No. 7, pp. 255-267, with 5 text figs.

1909 DOFLEIN, F. Lehrbuch der Protozoenkunde. Eine Darstellung der Naturgeschichte der Protozoen mit besonderer Berücksichtigung der parasitischen und pathogenen Formen. 2. Auflage der "Protozoen als Parasiten und Krankheitserreger." Mit 825 Abbildungen im Text, 914 pp., 8 vols. Jena, Gustav Fischer, Leishmania, pp. 701-704; Leishmaniosen, pp. 755; Schriften über Leishmanien, pp. 827-828.

1911 —— Ditto. (Dritte stark vermehrte Auflage. Mit 951 Abbildungen im Text). Jena, Gustav Fischer.

1903 DONOVAN, C. On the possibility of the occurrence of trypanosomiasis in India. Brit. Med. Journ., July 11, p. 79. Lancet, July 11, p. 44.

1903 —— A possible cause of kala azar. Ind. Med. Gazette, December, vol. 38, p. 478.

1903 —— The etiology of one of the heterogeneous fevers of India. Brit. Med. Journ., vol. 2, p. 1401.

1904 —— Piroplasmosis. A history of the discovery of the Donovan bodies in Madras. Ind. Med. Gazette, vol. 39, pp. 321-327.

1904 —— Human piroplasmosis. The Lancet, Sept. 10, vol. 2, pp. 744-750. 1905, January 21, pp. 155-156. Brit. Med. Journ., Sept. 17, vol. 2, pp. 651-653 (Abstract). Journ. Trop. Med. and Hyg., No. 15, p. 258.

1905 —— Medical cases from Madras General Hospital. Ind. Med. Gazette, November, vol. 40, p. 412.

1909 —— Kala-azar. Annual Report and Statistics of the Government General Hospital, Madras, for the year 1908. Issued from the Government Press, Madras, pp. 28-32.

1909 —— Notes on malaria and kala-azar. Journ. Trop. Med. and Hyg., July 1, vol. 12, No. 13, pp. 198-201.

1909 —— Kala-azar in Madras, especially with regard to its connection with the dog and the bug (*Conorrhinus*). Lancet, Nov. 20, vol. 2, pp. 1405-1406.

1909 —— Kala-azar in Madras. Supplement to the Ind. Med. Gaz., March, No. 44, pp. 16-8; Trans. Bombay Med. Congress, Feb. 24, pp. 159-160.

1913 —— Kala-azar, its distribution and the probable modes of infection. Ind. Journ. Med. Res., July, vol. 1, No. 1, pp. 177-184. Proceedings 3rd meeting General Mal. Com. held at Madras, November, 1912, pp. 211-214. 1913, Ind. Med. Rec., July, vol. 33, No. 7, pp. 156-157.

1910 DONZELLO. Sull' anemia splenica infantile da parassiti di Leishman (kala-azar?). Il Policlinico (sezione practica), March 20, vol. 17, No. 12, pp. 368-370.

1900 DOPTER, M. C. Les dysenteries. Étude bactériologique. Bibliothèque de microbiologie et de parasitologie, pp. 209, 8vo. Calmette, A., and Bezançon, F., Dysent. Ersch. bei Kala-azar, Chap. V.

1925 DOSTROWSKY, A. Ueber einen neuen endemischen Leishmania-herd in Palastina. Arch. f. Schiffs u. Trop.-Hyg., March 1, vol. 29, No. 3, pp. 101-111, with 4 text figs.

1925 DRBOHLAV, J. J. Studies on the relation of insect herpetomonad and crithidial flagellates to leishmaniasis. I.—Inoculation of mammals with insect flagellates. II.—The specificity of the various insect flagellates for certain hosts, as indicated by cross infection experiments. III.—Cultivation requirements. Amer. Journ. Hyg., September, vol. 5, pp. 580-507, pp. 509-610, pp. 611-621.

1900 DSCHUNKOWSKY and LUHS, J. Leishmania beim Hunde in Transkaukasien. 9e Cong. internat. de Med. Veterin., La Haye, September.

1926 DUDGEON, L. S. Case of kala-azar in a child treated with intravenous sodium antimony tartrate. Death. Microscopical examination of the various viscera. Proc. Roy. Soc. Med., vol 19 (Sect. of Trop. Dis. and Parasit.), pp. 11-13.

1922 DUNBAR, B. H. V. Notes on a (kala-azar) case treated by intravenous injections of tartar emetic. Journ. Roy. Army Med. Corps, October, vol. 39, No. 4, pp. 298-300.

1924 DUNCAN, J. T., and MANSON BAHR, P. H. The action of "Bayer 205" on the tissues in fatal cases of kala-azar and trypanosomiasis. Proceedings of a Laboratory Meeting of the Society held at the London School of Tropical Medicine, on December 13, 1923. Trans. Roy. Soc. Trop. Med. Hyg., vol. 17, Nos. 6 and 7, pp. 392-402.

1926 DUNLAP, A. M. Chronic laryngeal stenosis complicating kala-azar and diphtheria. China Med. Journ., May, vol. 40, pp. 409-415.

1922 DUTT, A. C. Treatment of kala azar. Ind. Med. Gazette, vol. 57, July, No. 7, p. 279.

1924 DUTT, MADAN MOHAN. Certain observations on cases of enlarged spleen as met with in Bengal. Cal. Med. Journ., June, vol. 18, No. 12, pp. 780-703.

1912 EASTON, P. G., Capt. R.A.M.C. Treatment of kala-azar. Journ. Roy. Army Med. Corps, April, vol. 18, No. 4, p. 491.

1912 Eightieth Annual Meeting of the British Medical Association held in Liverpool, July 19 to 22. Section of Tropical Medicine. Papers dealing with leishmaniasis. Brit. Med. Journ., vol. 2.

1900 ELDERS, C. Kala-azar in Deli. Geneeskundig Tijdschrift voor Nederlandsch-Indië, Batavia, vol. 99, afl. 6, pp. 785-789.

1910 —— Leishmaniasis acuta (kala-azar) bij een Javaan op Sumatra. Geneeskundig Tijdschrift voor Nederlandsch-Indië, afl. 2, pp. 193-190.

1911 —— Über eine klinisch und ätiologisch der Trypanosomiasis und Schlafkrankheit verwandte Krankheit bei Javanen auf Sumatra. Archiv für Schiffs- und Tropen-Hygiene, vol. 15, Part 1, pp. 1-7.

1904 ELLIS, A. G. The Leishman-Donovan body. Amer. Med., vol. 8, No. 21, p. 901.

1920 ELWES, F. F. Antimony in kala-azar. Brit. Med. Journ., Oct. 16, p. 610.

1924 —— MENON, V. K. N., and RAMAKRISHNAN, P. S. The formolgel (aldehyde) test as a means of diagnosis of kala azar. Ind. Med. Gazette, April, vol. 59, No. 4, pp. 175-177.

1911 ENDE. Kala-azar in Japan. Medizinische Gesellschaft in Tokio, Sessions January-April. Reprinted in Deutsche medizinische Wochenschrift, No. 29, p. 1376.

1900 ENSOR, H. The treatment of kala-azar by the use of senega. Journ. Roy. Army Med. Corps, December, vol. 13, No. 6, pp. 667-672.

1926 ESCHBACH, H. Kala-azar in an adult. Bull. et Mém. Soc. Méd. Hôp. de Paris, Nov. 18, year 42, 3rd series, vol. 50, No. 34, pp. 1568-1570.

1914 ESCOMEL, EDMUNDO. Leishmania flagelada en el Peru. Crónica Medica, Lima, July 15, vol. 31, No. 613, pp. 224-227.

1925 —— Leishmaniose américaine des sinus frontaux. Traitement favorable par l'iodure double de quinine et bismuth. Bull. Soc. Path. Exot., vol. 18, pp. 634-639. Previously published in Spanish in Cronica Méd., Lima, vol. 13, pp. 129-132.

1892 EVANS J. F. A note on the pathology of kala-azar or beri-beri of Assam. Ind. Med. Gazette, vol. 27, pp. 330-332 and pp. 353-354.

1923 FABER, H. K., and SCHUSSLER, Jr., H. Leishmaniasis in the United States. Rep. third Am. case of kala-azar. Journ. Amer. Med. Assoc., January 13, vol. 80, No. 2, pp. 93-97.

1915 FAGIUOLI, ANTONIO. Contributo clinico ed anatomo-patologico allo studio dell' anæmia da leishmania. Riforma Med., Oct. 23, vol. 30, No. 43, pp. 1188-1191; Oct. 30, No. 44, pp. 1214-1217.

1912 FANTHAM, H. B. Some insect flagellates and the problem of the transmission of leishmania. Brit. Med. Journ., Nov. 2, pp. 1196-1197.

1915 —— Herpetomonads and vertebrates: A correction of a recent contribution on "Leishmania problems." Journ. Trop. Med. and Hyg., Dec. 15, vol. 18, No. 24, pp. 277-281.

1921 FARGHER, R. G., and GRAY, W. H. The chemotherapy of antimony. Journ. Pharm. Exp. Therapy, vol. 18, p. 341.

1909 FELETTI, R. Il kala-azar a Catania. La Riforma Medica, Dec. 13, vol. 25, No. 50, pp. 1373 1375.

1910 —— Contribuzione allo studio delle leishmanie. Pathologica, March 1, vol. 2, No. 32, p. 103.

1910 —— Sul kala azar osservato a Catania. Atti dell' Accademia Gioenia di scienze naturali, Catania, section 5, vol. 3. Reprinted in Virchow's Jahresbericht, p. 534.

1926 —— Lo stovarsolo nella cura del kala azar. Gaz. Internaz. Med.-Chir., vol. 31, pp. 111-114.

1915 FERNANDEZ, MARTINEZ F. Las leishmaniosis patogenas en el mediodia de España. Clin. Mod. Zaragoza, vol. 14, pp. 149 and 189.

1914 FIEBER. Kurven bei der kindlichen Leishmaniosis. Deut. Arch. f. klin. Med., Jan. 31, vol. 113, Nos. 3-4, pp. 354-371.

1897 FINK, G. H. The so-called kala-azar of Assam. Ind. Med. Gazette, vol. 32, pp. 214 216, 248-252, 293-298. Abstract in Journ. Trop. Med., 1905, vol. 8, p. 156.

1899 —— Kala-azar and ankylostomiasis in Assam. Ind. Med. Rec., vol. 16, pp. 775-790.

1916 FINZI, G. Leishmaniose et tuberculose chez le chien. Bull. Soc. Path. Exot., July, vol. 9, No. 7, pp. 429-432.

1907 FLORENCE. Splénomégalie tropicale de Donovan. Toulouse médical, série 2, vol. 9, pp. 253-256.

1923 FONZO, F. Kala-azar e malaria in raporto alla stagione e al clima. Pediatria, Dec. 1, vol. 31, No. 23, pp. 1262-1269.

1924 FOSTER, P. Urea stibamine in the treatment of kala-azar under tea garden conditions. Proc. Assam Branch, British Medical Association, annual meeting, Jorhat, February 16-17, 1924, pp. 24 26. Ind. Med. Gazette, August, vol. 59, No. 8, pp. 391-393.

1926 —— A note on the control of kala-azar outbreaks by means of therapeutic measures. Proc. Assam Branch Brit. Med. Assoc. Annual Meeting, Silchar, March 1 and 2, pp. 17-18.

1921 FOTI, P., and JAVARONE, N. Contributo statistico clinico alla terapia specifica della leishmaniosi interna. Pediatria, Feb. 15, vol. 29, No. 4, pp. 145-155.

1921 FOX, E. C. R., and MACKIE, F. P. The formolgel test in kala-azar. Ind. Med. Gazette, October, vol. 56, No. 10, pp. 374-375.

1926 FRANCA, CARLOS. Thoughts on the leishmanias. Mém. et Études du Museum Zool. de l'Univ. de Coimbra, ser. 2, No. 2, p. 7.

1910 FRANCHINI, G. Lipuria in un caso di kala-azar complicato da anchilostomiasi. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia. Part 2, pp. 30-40. Tipografia Labicana.

1911 —— Kultur und Modalitäten der Entwicklung der Leishman-Donovan'schen Körperchen. Berliner klinische Wochenschr., May 15, vol. 48, No. 20, pp. 896-897.

1911 —— La vita e lo sviluppo della *Leishmania donovani* nelle cimici nelle pulci e nei pidocchi. Malaria e Malattie dei Paesi Caldi, Part 4. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da U. Gabbi, Part 2. Tipogr.: S. Guerriera, Messina.

1911 —— Sulla resistenza della *Leishmania donovani* alle varie temperature. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da U. Gabbi, Part 2. Tipogr.: S. Guerriera, Messina.

1911 —— Infezione sperimentale da *Leishmania donovani* nella Cavia. Pathologica, June 10, vol. 3, No. 62, pp. 255-256.

1911 —— Note on leishmania and mosquito. Lancet, Nov. 4, vol. 2, pp. 1268-1269.

1911 —— Ancora sulla morfologica della *Leishmania donovani* in cultura e sul suo rapporto col globulo rosso. Mal. e Malat. dei Paesi Caldi, December, vol. 2, No. 12, pp. 349-352.

1911 —— Su di alcune rarissime forme di *Leishmania donovani* osservate in tubi vecchi di culture. Mal. e Malat. dei Paesi Caldi, December, vol. 2, No. 12, pp. 352-354.

1911 —— Leishmania and mosquitoes. (Correspondence.) Lancet, December 23, vol. 2, No. 4608, p. 1801.
1911 —— Sulla resistenza della *Leishmania donovani* alle varie temperature. Mal. e Malat. dei Paesi Caldi, August, vol. 2, No. 8, pp. 227-229.
1911 —— Sulla morfologia e sul ciclo di sviluppo della *Leishmania donovani* nelle culture. Mal. e Malat. dei Paesi Caldi, Rome, September, vol. 2, No. 9, pp. 253-267.
1911 —— Histologische Veränderungen und parasitärer Befund bei einem an Infektion durch *Leishmania donovani* verendeten Meerschweinchen. Münchener med. Wochenschr., Sept. 26, vol. 58, No. 29, p. 2067.
1911 —— La *Leishmania donovani* puo vivere e svilupparsi nel tubo digerente dell' anopheles. Nota preventiva. Pathologica, Nov. 1, vol. 3, No. 72, pp. 611-613.
1911 —— La *Leishmania donovani* puo vivere e svilupparsi nel tubo digerente dell' anopheles cl. Mal. e Malat. dei Paesi Caldi, Rome, November, vol. 2, No. 11, pp. 324-326.
1912 —— Leishmania e zanzare. Ulteriori esperienze con zanzare e parassiti splenici. Riforma Medica, Dec. 7, vol. 28, No. 49, pp. 1355-1358.
1912 —— Leishmania et punaises. Bull. Soc. Path. Exot., December, vol. 5, No. 10, pp. 817-819.
1912 —— Leishmania and mosquitoes. (Correspondence.) The Lancet, Feb. 24, vol. 1, No. 4617, p. 534.
1912 —— On the presence of leishmania in the digestive tract of *Anopheles maculipennis*. From the Analytical Laboratory of the Ospedale Maggiore, Bologna, under the Directorship of Prof. G. Vannini. Annals of Trop. Med. and Parasitology, May 29, series T. M., vol. 6, No. 1B, pp. 41-52.
1912 —— A proposito dell' articolo del Dr. Franchini in Pathologica num. 72 (vidi anche num. 74, 78, 81). (Editorial.) Pathologica, July 1, vol. 4, No. 88, p. 408.
1912 —— Leishmanie e zanzare. Riforma Medica, Sept. 7, vol. 28, No. 36, pp. 981-982.
1911 FRANCO, E. E. Intorno ad un caso di kala-azar. A Medicina Contemporanea. Lisbon, September.
1920 —— Hemohistoblastes et leurs dérivés monocytiques, lymphocytiques et granulocytiques dans la rate et dans le sang circulant d'enfants affectés de leishmaniose. C. R. Soc. Biol., July, vol. 83, No. 26, pp. 1187-1189.
1921 —— Sulle leishmaniosi. Relazione al Congresso Luso. Spagnuolo di Porto, June. Reprinted from La Med. Ital., Feb. 28, No. 2, p. 4.
1921 —— Études sur les leishmanioses. Travaux de l'Institut de Pathologie Générale et d'Anatomie Pathologique de l'Université de Lisbonne. Journ. Sci. Mat. Fis. e Naturalis, series 3, No. 8, p. 8.
1922 —— Anatomia patologica della leishmaniosi infantile. Arch. Portugaises des Sci. Biologiques, Lisbon, vol. 1, No. 1, pp. 31-95.
1922 —— Leishmaniosi viscerale dell' adulto (kala-azar). Pathologica, April 1, vol. 14, No. 321, pp. 193-196.
1922 —— Le alterazioni spleniche nella leishmaniosi infantile. Journ. de Sci. Mat. Fisicas, e Naturais, series 3, No. 11, p. 40.
1922 —— Particularités de culture de certains flagellés. De la culture des leishmania dans le milieu de Yoshida. Bull. Soc. Path. Exot., July 12, vol. 15, pp. 551-555.
1922 —— Hémohistoblastes et leurs dérivés monocytiques, lymphocytiques et granulocytiques dans la rate et dans le sang circulant d'enfants atteints de leishmaniose. Reprinted from Journ. Sci. Mat. Fisc. e Nat., Lisbon, No. 9, p. 6.
1915 FRANKEL. Die Auzentimittel Synthesie.
1905 FREER, G. D. Two fatal cases of kala-azar, admitted to the Penang General Hospital, in whom Leishman-Donovan bodies were found. Journ. of the Malaya Branch of the British Med. Assoc., Singapore, 1905-6, new series, No. 2, pp. 69-72.
1918 FRIAS, A. Kala-azar on Mediterranean coast of Spain. Arch. Españoles de Pediatria, June, vol. 2, No. 8, p. 321 (quoted in Journ. Amer. Med. Assoc., vol. 71, pp. 1782).
1918 —— and ROIG. El kala-azar infantil en Reus y su Camarca. Arch. Españoles de Pediatria, June, vol. 2, No. 8, pp. 321-340.
1911 FULCI, FRANCESCO and BASILE, CARLO. Un caso di kala-azar a Roma. Atti della reale Accademia dei Lincei, Rendiconti, Jan. 22, serie 5, 1° semestre, vol. 2, pp. 132-136. Clinica ostetrica, vol. 13, p. 88.
1906 FÜLLEBORN, FRIEDRICH. Über Kala-azar oder tropische Splenomegalie. Archiv für Schiffs- und Tropen-Hygiene, vol. 10, No. 24, pp. 766-776.
1907 —— Kala-azar. Biol. Abt. ärztl. Ver. Münch. med. Wochenschr., Jahrg. 54, p. 442.

1907 FÜLLEBORN, FRIEDRICH. Über die Kala-azar (tropische Splenomegalie) genannte Krankheit; mit Demonstration von Präparaten. Verhandlungen Ges. Deutscher Naturforscher und Ärzte Versammlung 78, Part 2, 2nd half, pp. 390-391.
1907 —— Kala-azar. Die Umschau, Frankfurt a. M., vol. 11, pp. 970-972.
1925 FUNAIOLI, G. La leishmaniosi canina in Tripolitania. Arch. Ital. Sci. Med Colon., vol. 6, pp. 12-14.
1925 FURUMORI, KAMEHIRA. On a case of kala-azar occurring in Tokyo. Chugwai Ija Shimpo. Central Med. Journ., January, vol. 44, No. 2. (Summarized in Japan Med. World, March 15, vol. 5, No. 3, p. 66.)
1909 FUSCO, G. Anemie spleniche e corpi di Leishman. Ricerche in corso. In report by G. Rummo, Riforma Medica, vol. 25, No. 27, p. 738.

1909 GABBI, UMBERTO. Focolai endemici della varietà febbrile dell' anemia splenica infettiva dei bambini. Prima communicazione. Il Policlinico, Medical Section, vol. 16, Part 1, pp. 1-5.
1909 —— Nuovo contributo clinico allo studio del kala-azar in Sicilia (terza comunicazione). Il Policlinico, sezione medica, Rome, vol. 16, fasc. 6, pp. 241-248.
1909 —— Nuovo contributo clinico allo studio del kala-azar in Sicilia. Bollettino delle cliniche, Milan, vol. 26, pp. 251-261.
1909 —— Nuovo contributo clinico allo studio del kala-azar. 11 pp., Napoli, Premiato stabilimento tipograpico F. Sangiovanni, Vico Salata ai Ventaglieri, 14. Cited in Archives de l'Institut Pasteur de Tunis, November, vol. 4, p. 209.
1910 —— Il kala-azar nella seconda infanzia nell' adolescenza e nell' adulto. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia, vol. 2, pp. 1-6. Tipografia Labicana. Atti della reale Accademia dei Lincei, Rendiconti, series 5, vol. 19, 1° semestre, pp. 407-408. Riforma Medica, vol. 26, No. 18, pp. 477-470.
1910 —— Intorno al kala-azar. Il focolaio endemico di Bordonaro. Studio clinico. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia. Sezione di malattie esotiche del R. Instituto di clinica medica di Roma. Rivista critica di clinica medica. Vol. 11, pp. 113-129.
1910 —— Kala-azar in Sicilia e Calabria. Studio intorno ad alcune malattie tropicali della Calabria e della Sicilia, vol. 1, pp. 1-47.
1910 —— Kala-azar in Italia. Sunti e rivista critica. Malaria e Malattie dei Paesi Caldi (formerly Malaria e Malattie affini), anno 1, No. 3, pp. 80-85; Nos. 4-5, pp. 118-131.
1910 —— La mobilità della milza nel kala-azar. Nota di semeiotica. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia, vol. 2, pp. 7-8. Roma: Tipografia Labicana.
1910 —— Intorno alla ricorrenza primaverile dei casi di kala-azar. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia, vol. 3, pp. 33-36. Roma: Tipografia F. Centenari. Riforma medica, August 28, vol. 26, No. 35, pp. 953-954.
1910 —— Le infezioni febbrile simulatrici della malaria. Studio intorno ad aucune malattie tropicali della Calabria e della Sicilia, vol. 3, pp. 46-49. Roma: Tipografia F. Centenari.
1910 —— Intorno al kala azar (Nuovi contributi). Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia, vol. 3, pp. 37-45. Roma: Tipografia F. Centenari. Riforma medica, November 21, vol. 26, No. 47, pp. 1298-1302.
1911 —— La patologia a clinica delle malattie tropicali nel loro contenuto scientifico e nella loro importanza practica, specie in reguardo a quelle dei paesi del bacino Mediterraneo. Gazetta Internationale di Medicina, Chirurgia Igiene, Interessi Professionali, Napoli, No. 6. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da Umberto Gabbi, pp. 1-20. Roma: Tipografia F. Centenari.
1911 —— La patologia tropicale dei paesi del bacino Mediterraneo nel suo contenuto scientifico e nella sua importanza practica. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da Umberto Gabbi, pp. 35-40. Roma: Tipografiia F. Centenari Pathologica, Jan. 15, vol. 3, No. 53, pp. 24-27.
1911 —— Le pulci canina et umana non propagano il kala-azar. Mal. e Malat. d. Paesi Caldi, Rome, October, vol. 2, No. 10, pp. 285-288.
1911 —— Note on tropical diseases in Southern Italy. Annals of Tropical Medicine and Parasitology, August 1, vol. 4, No. 2, pp. 135-138.
1911 —— Intorno alla ricorrenza primaverile del kala-azar. Mal. e Malat. dei Paesi Caldi, Rome, October, vol. 2, No. 10, pp. 288-290.
1911 —— (1) Le pulci canine ed umana non propagano il kala-azar. (2) Sulla ricorrenza primaverile el kala-azar. (1° Congresso Internazionale dei Patalogie, October 2-5. Rendiconto originale.) Pathologica, December 1, vol. 3, No. 74, pp. 680-681.

1911 —— Il ponos e kala-azar. Atti della reale Accademia dei Lincie, Rendiconti, Rome, 5 serie, vol. 20, 1° semestre, fasc. 3, pp. 187-188.
1912 —— Reporto di leishmania nell' essudato di una stomatite ulcerosa complicante un caso di kala-azar. Mal. e Malat. dei Paesi Caldi, March, vol. 3, No. 3, pp. 78-79.
1912 —— Leishmania and mosquitoes. (Correspondence.) The Lancet, Feb. 24, vol. 182, No. 4617, p. 534.
1912 —— Sulla identità delle leishmania infantum e donovani. Mal. e Malat. d. Paesi Caldi, December, vol. 3, No. 12, pp. 334-335.
1912 —— Il kala-azar infantile ed a leishmania infantum al lume delle ultime ricerche. Mal. e Malat. d. Paesi Caldi, December, vol. 3, No. 12, pp. 336-344.
1913 —— Sulla identità clinica et etiologica della leishmaniosi umana e canina. Pathologica, Sept. 15, vol. 5, No. 117, pp. 543-552.
1913 —— On the identity of infantile and Donovan's leishmania (kala-azar). Journ. Trop. Med. and Hygiene, vol. 16, July 1, No. 13, pp. 198 199.
1913 —— Sulla storia del kala-azar del Mediterraneo. Mal. et Malat. d. Paesi Caldi, April-May, vol. 4, No. 3, pp. 198-202.
1913 —— Intorno all'origine canina della leishmaniosi interna (kala-azar). Mal. e Malat. d. Paesi Caldi, Jan., vol. 4, No. 1, pp. 7-19.
1914 —— Il kala-azar Indiano e Mediterraneo sono Identici. Nuove indagini sperimentali. Mal. e Malat. d. Paesi Caldi, Feb., vol. 5, No. 1, pp. 14-22, Pathologica, Feb. 1, vol. 6, No. 126, pp. 69-74.
1913 —— Über den Ursprung der Leishmaniosis interna (kala-azar) vom Hunde. Centralbl. f. Bakt., 1. Abt. Orig., July 3, vol. 69, No. 7, pp. 504-516.
1913 —— Au sujet de l'historique du kala azar Mediterranéen. Bull. Soc. Path. Exot., March, vol. 6, No. 3, pp. 141-143.
1913 —— Leishmaniosi umana e metodi di polemica. Mal. e Malat. d. Paesi Caldi, June-July, vol. 4, No. 4, pp. 269-270.
1914 —— Le complicanze della leishmaniosi interna. Lavori d Soc. Italiana di Patologia Esotica, pp. 75-77.
1914 —— Über das Auftreten der Leishmaniosis interna (Kala-azar) in bestimmten Jahreszeiten. Beiheft. z. Arch. f. Schiffs- u. Trop.-Hyg., vol. 18, No. 7, pp. 83-87.
1914 —— Le complicanze della bocca e dell' orecchio nella leishmaniosi. Lavori d. Soc. Italiana di Patologia Esotica, pp. 77-82.
1914 —— Le complicazioni del sangue e degli organi emopoietici. Lavori d. Soc. Italiana di Patalogia Esotica, pp. 105-107.
1914 —— Experimentelle Infektion indischer Hunde durch das "Virus der Mittelmeer Kala-azar." Beiheft z. Arch. f. Schiffs- u. Trop.-Hyg., vol. 18, No. 7, pp. 79 82.
1917 —— Sulla unicità etiologica delle varie leishmaniosi. Mal. e Malat. d. Paesi Caldi, Jan.-Feb., vol. 8, No. 1, pp. 10-20.
1923 —— Kala-azar oder Innere Leishmaniose, Handbuch der Tropenkrankheiten herausgegeben von Prof. Dr. Carl Mense, Kassel, Zweite auflage, Vierter Band.
1909 —— and CARACCIOLO, R. Über Kala-azar in Sicilien und Calabrien. Centralblatt für Bakteriologie, 1 Abt. Originale 1, Part 4, pp. 424-427.
1913 —— LOMBARDO, P. P., and MONTORO, G. Inchiesta intorno al kala-azar nelle Provincie della Sicilia orientale e della Calabria. Inf. resultati raggiunti. Mal. d. Malat. d. Paesi Caldi, June-July, vol. 4, No. 4, pp. 230-253.
1914 —— PELLEGRINO, P. L., and MONTORO, G. Untersuchung über die Kala-azar in den östlichen Provinzen Siziliens und Unter-Kalabriens, sowie über die erzielten Resultate. Zentralbl. f. Bakt., 1 Abt. Orig., Jan. 24, vol. 72, Nos. 6-7, pp. 505-516.
1910 —— and SCORDO, F. Infezione melitense dell' uomo e delle capre in alcune citta delle coste della Grecia. Il ponos e kala-azar. Mal. e Malat. del Paesi Caldi, 1., Part 8. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da U. Gabbi, vol. 2. Tipogr. : S. Guerriera, Messina.
1910 —— and VISENTINI, A. Il kala-azar Italiano e trasmissibile al cane. Pathologica, April 1, vol. 2, No. 34, pp. 149-150.
1910 DE GAETANI-GIUNTA, G. L'anaemia splenica infantile febbrile. Gazzetta degli Ospedali, No. 15, pp. 153-154.
1911 GALLE, Prof. Is ponos kala-azar? Prof. Galle's discovery (abstract). Journ. Trop. Med. and Hyg., Jan. 2, vol. 14, No. 1, p. 9.
1925 GALLO, CARMINE. Ricerche sul contenuto in ferro del sangue nelle anemie spleniche e nella leishmaniosi infantile. Pediatria, March 1, vol. 33, No. 5, pp 251-256.
1922 GANGULI, L. Formolgel test in kala-azar. Ind. Med. Gazette, Feb., vol. 57, No. 2, p. 77.

1924 GANGULI, P. Investigations on the ætiology and treatment of kala-azar and some other common diseases in Bengal. Ind. Med. Rec., February, vol. 43, No. 2, pp. 34-36.
1925 —— The value of the aldehyde test in the diagnosis of kala-azar. Ind. Med. Gazette, May, vol. 60, pp. 204-206.
1925 —— Some investigations into the biochemistry of blood and other factors of kala-azar patients. Ind. Journ. Med., vol 6, October, pp. 12-13.
1914 GARCIA DEL DISETRO, J. El primero caso de kala-azar en Madrid. Rev. Clin. de Madrid, vol. 12, pp. 191-197.
1922 GATÉ, J., and PAPACOSTAS, G. La formol-gelification des serums dans diverses maladies. C. R. Soc. Biol., July, vol. 87, No. 26, pp. 543-544.
1921 GATT, T. E. H. A case of splenic anæmia in a child due to *Leishmania infantum*. Roy. Army Med. Corps, August, vol. 37, No. 2, pp. 142-143.
1914 GATTI, G. Nuovi casi di leishmaniosi infantile osservati a Napoli e dintorni. Pediatria, November, vol. 22, No. 11, pp. 834-849.
1926 —— and COOKE, W. E. A case of congenital kala-azar. Lancet, Dec. 11, p. 1209.
1923 GAZDER, J. D. Sidelights on the treatment and prevention of kala-azar and malaria in Bengal. Ind. Med. Record, August, pp. 215-216.
1925 GERARD, F., and SALVO, CORTESI. Nouvelles observations de kala-azar en Tunisie. Arch. Inst. Pasteur de Tunis, December, vol. 14, No. 4, pp. 462-464.
1924 GERSCHENOWITSCH, R. S. On the comparative value of existing methods of treatment of infantile leishmaniosis. Russian Journ. of Trop. Med., No. 2, pp. 11-16.
1916 GHOSH, HARINATH. The speedy recovery of a case of kala-azar by intravenous injection of sodium antimony tartrate with sodium cinnamate and berberine hydrochloride. Calcutta Med. Journ., Jan. 6, p. 6.
1916 —— Further reports of recovery of cases of kala azar by intravenous injection of a compound solution of sodium antimony tartrate. Calcutta Med. Journ., October, pp. 97 104.
1923 —— Remarks on anti malarial and anti-kala-azar works. Ind. Med. Record, September, pp. 247-251.
1925 GHOSH, N. L. A new factor in the control of kala-azar. Ind. Med. Rec., vol. 45, pp. 4-6.
1923 GHOSH, N. N. Some observations on the treatment of kala azar. Ind. Journ. Med., pp. 201-205.
1892 GIANTURCO, V., and PIANESE, G. Ricerche batteriologiche sperimentali e istologiche, in un caso di pseudoleucemia infantile infettiva (Cardarelli) o anemia splenica (Fede). Gazetta delle Cliniche, vol. 3, pp. 305-310.
1890 GILES, GEORGE M. Report of an investigation into the causes of the diseases known in Assam as kala-azar and beri-beri. 156 pp. Assam Secretarial Press.
1892 —— Notes on anchylostomiasis, being for the most part a résumé of a report on the diseases known in Assam as kala-azar and beri-beri. Ind. Med. Gazette, vol. 27, pp. 170-173, and 193-196.
1898 —— The etiology of kala-azar. Ind. Med. Gazette, vol. 33, No. 1, pp. 1-4.
1898 —— The etiology of kala-azar: Dr. Giles' criticism of Dr. Rogers' report. Ind. Med. Record, vol. 14, pp. 35-38.
1898 —— Kala-azar. Brit. Med. Journ., vol. 1, p. 861.
1899 —— Alleged infectiousness of malaria: kala-azar. Ind. Med. Gazette, vol. 34, p. 37.
1915 DI GIORGIO, G. A proposito di 41 casi di leishmaniosi infantile osservati nella clinica pediatrica di Palermo durante l'anno scolastico, 1912-1913. Gaz. Internaz. Med. Chirurg. Igiene, Jan. 9, vol. 1, pp. 4-10; Jan. 16, No. 2, pp. 24-28.
1918 GIOSEFFI TRIEST, M. Ein Fall von Leishmaniosis. Kala-azar. Münch. med Woch., August 13, vol. 65, No. 33, pp. 910 911.
1926 GIRAUD, P. Le kala-azar infantile en France. (Étude épidémiologique et thérapeutique.) Arch. de Méd. d. Enf., vol. 19, pp. 185-208.
1925 —— and CAUDIERE, M. Sur l'anatomie pathologique du kala-azar infantile; étude anatomique de quatre cas autochtones. Ann. d'Anat. Path. Méd.-Chir., April 9, vol. 2, pp. 153-168.
1926 —— —— Les lesions histologiques du kala-azar infantile. C. R. Soc. Biol., vol. 94, pp. 885-887.
1924 —— and ZUCARELLI, J. Le kala-azar infantile autochtone dans la région Mediterranéenne. Paris Méd., March 22, No. 12, vol. 14, pp. 278-281.
1907 GIRAULT, A. ARSENE. The Indian bed-bug and the kala-azar disease. Science, New York. New series. Vol. 25, p. 1004.
1926 GIUFFRE, M. La reazione meiostogminica di Ascoli nella leishmaniosi interna. Pediatria, vol. 34, pp. 337-349.

1914 GIUGNI, FRANCESCO. Le complicazioni renali del kala-azar. Lavori de Soc. Italiana di Patalogia Esotica, pp. 92-104.

1914 —— La emocultura della *Leishmania donovani* dal sangue periferico in un caso di kala-azar nel bacino del Mediterraneo. Pathologica, June 1, vol. 6, No. 134, pp. 284-285.

1914 —— Ricerche sulla vitalità e lo sviluppo della *Leishmania donovani* nei terreni culturali. Mal. e Malat. d. Paesi Caldi, May-June, vol. 5, No. 3, pp. 156-161.

1915 —— Sulla presenza della *Leishmania donovani* e lo sviluppo culturale del sangue periferico nel kala-azar. Pathologica, vol. 7, No. 151, pp. 84-87, and Mal. e Malat. d. Paesi Caldi, Jan.-Feb., vol. 6, No. 1, pp. 16-20.

1915 —— Alcuni tentativi di transmissione della leishmaniosi canina. Adamento clinico e dati necroscopici di un caso di leishmaniosi nel cane. Mal. e Malat. d. Paesi Caldi, March-April, vol. 6, No. 2, pp. 77-81.

1915 —— and BENONI, J. Sul comportamento in vitro delle leishmania col melitense, coi germi tifici, paratifici, e coi loro agglutinanti. La contemporanea infezione di febre Mediterranea e leishmania int. Mal. e Malat. d. Paesi Caldi, March-April, vol. 6, No. 2, pp. 89-94.

1912 GOERE, DR. Leishmaniasis. Leçon faite à l'Institut Pasteur par M. le Professeur A. Laveran, recueillie par M. le Dr. Goere. Archives de Méd. et Phar. navales, Jan., vol. 97, No. 1, pp. 43-52.

1913 GORETTI, G. Su di una forma rara di splenomegalia. Contributo anatomo patologico. Mal. e Malat. d. Paesi Caldi, March, vol. 4, No. 2, pp. 117-122.

1925 GRAHAM, J. D. La commission du kala-azar dans l'Inde Britannique. Bull. Office Internat. d'Hyg. Publique, vol. 17, pp. 512-516.

1907 GRANGER, T. A. Leishman-Donovan infection in a Gurkha. Ind. Med. Gazette, vol. 42, pp. 13-14.

1913 GRAY, A. C. H. Report on some observations made and work done at the Pasteur Institute, Tunis. Journ. Roy. Army Med. Corps, December, vol. 21, No. 6, pp. 696-712.

1913 —— Leishmaniose naturelle du chien à Tunis. Bull. Soc. Path. Exot., March, vol. 6, No. 3, pp. 165-166.

1927 GRAZIANO, FRANCESCO. Nuovo contributo all terapia del kala-azar (Treatment of kala azar). Pediatria, April 1, vol. 35, No. 7, pp. 359-364.

1925 GREIG, E. D. W., and KUNDU, S. Observations on the quantity of urea stibamine, "471" von Heyden, and stibamine glucoside required for the complete treatment of a case of kala azar. Ind. Journ. Med. Res., April, vol. 12, No. 4, pp. 679-688.

1925 —— —— Observations on two cases of kala-azar resistant to treatment. Ind. Journ. Med. Res., April, vol. 12, No. 4, pp. 689-694.

1925 —— —— Observations of glycosuria and blood sugar content in kala-azar. Ind Journ. Med. Res., April, vol. 12, No. 4, pp. 695-699.

1925 —— and CHRISTOPHERS, S. R. Infection of a monkey (*Macacus rhesus*) with the parasite of Indian kala-azar following the introduction of infective material into the lumen of the small intestine. Ind. Journ. Med. Res., July, vol. 13, pp. 151-157.

1916 GUIDI, G. Terapia della leishmaniosi interna. Rev. Clin. Pediatria, vol. 14, No. 5, pp. 265-270.

1904 GUITERAS, JUAN. Esplenomegalia tropical; el nuevo parasito de Leishman y Donovan: revista de la literatura con algunas observaciones. Revista de medicina tropica, vol. 5, pp. 69-76.

1925 GUPTA, P. N. Kala-azar. Ind. Med. Rec., vol. 45, pp. 319-325.

1926 GUPTA, N. R. An unusual form of dermal leishmaniasis. Ind. Journ. Med., September, vol. 7, pp. 133-135.

1913 GURKO, A. G. Vier Fälle von Kala azar. Zeitschr. f. Hyg. u. Infektionskr., May 20, vol. 74, No. 2, pp. 355-368.

1926 GURUWAMI MUDALIER, M. R., RAMAN, T. K., and RAMAN MENON, K. Kala-azar on the West Coast of India. Ind. Journ. Med. Res., January, vol. 13, No. 3, pp. 531-532.

1910 HAMIL, PHILIP. Intravenous injection of antimony tartrate in kala-azar. Brit. Med. Journ., July 5, p. 28.

1924 HANCE, J. B. The geographical distribution of kala-azar. Ind. Med. Gazette, February, vol. 59, No. 2, p. 82.

1910 HARRISON, J. HUGH. Kala-azar? Case treated in the British Honduras Hospital. Journ. Trop. Med. and Hyg., vol. 13, p. 76.

1909 HARRISON, L. W. A case of kala-azar. Journ. Roy. Army Med. Corps, vol. 12, pp. 63-64.

1910 HARRISON, W. S., and CUMMING, C. C. Two cases of kala-azar treated by the arylarsonates. Journ. Roy. Army Med. Corps, February, vol. 14, No. 2, pp. 200-204.
1907 HARRISS, S. A. Short report on four cases of Leishman-Donovan infection in Gurkhas. Ind. Med. Gazette, vol. 42, pp. 293-297.
1923 HARTMANN-KEPPEL, G. L. L'appendicite à leishmania. Presse Méd., March 28, No. 25, pp. 291-292.
1923 —— Un modo d'inoculation de la leishmaniose cutanée. Rev. Méd. et Hyg. Trop., July-August, vol. 15, No. 4, pp. 135-138.
1910 HARTWIG, G. Über den Einfluss einer chronischen Infektionskrankheit auf den Verlauf der Anemia splenica infantum; ein Beitrag zur Therapie dieses Leidens. Therapeutische Monatshefte, October, vol. 24, Part 10, pp. 527-530.
1916 HECKENROTH, F. Deux nouveaux cas de leishmaniose canine a Dakar. Bull. Soc. Path. Exot., November, vol. 9, No. 9, pp. 696-697.
1904 HEWLETT, R. T. The new parasite of kala-azar. Journ. of State Medicine, October, vol. 12, pp. 619-621.
1913 HILL, R. A. P. Note on a new sign in kala-azar. Lancet, August 9, p. 392.
1926 HINDLE, EDWARD, and PATTON, W. S. Experiments bearing on the susceptibility of the striped hamster (*Cricetulus griseus*) to leishmania of Chinese kala-azar. Proc. Roy. Soc., October 1, ser. B., vol. 100, No. B, 704, pp. 374-379.
1927 —— —— Sand-flies and Chinese kala-azar. (Correspondence.) Nature, March 26, vol. 119, No. 2995, p. 460.
1926 —— —— Resistance of leishmania cultures to cold. Proc. Roy. Soc., October 1, ser. B, vol. 100, No. B, 704, pp. 385-386.
1927 —— —— Sandflies and Chinese kala-azar (Correspondence). Nature, March 26, vol. 119, No. 2995, p. 460.
1926 —— HOU, P. C., and PATTON, W. S. Serological studies on Chinese kala azar. Proc. Roy. Soc., October 1, ser. B, vol. 100, No. B, 704, pp. 368-373.
1926 —— —— —— Reports from the Royal Society's Kala-azar Commission in China. Nos. 1 to 5. Proc. Roy. Soc., series B, vol. 100, pp. 368-390.
1926 HITTI, JOSEPH K. Infantile kala-azar in Syria. Journ. Amer. Med. Assoc., Oct. 16, vol. 87, No. 16, p. 1302.
1927 —— VARDON, A. C., and ZORAWAR SINGH. A preliminary note on a quick and simple test for the differentiation of malaria from kala-azar, enteric and other fevers. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 779-784.
1926 HODGSON, E. C. Clinical and pathological notes on cases of kala-azar. Proc. Assam Branch Brit. Med. Assoc. Annual Meeting, Silchar, March 1 and 2, pp. 11-16.
1907 HOLCOMB. The bed-bug as the intermediary host of Leishman-Donovan body of kala-azar. United States Naval Medical Bulletin, October 3, vol. 1, No. 3.
1905 HUNTER, W. K. Specimens of the Leishman-Donovan bodies. Glasgow Medico-Chirurgical Society. Glasgow Med. Journ., vol. 64, pp. 436-438.
1904 HUTCHINSON, JONATHAN. Discussion on the Leishman-Donovan body. Brit. Med. Journ., vol. 2, p. 658.

1924 IGLESIAS, GARZA, T. Una endemia grave de leishmaniosis infantil en España. Siglo Méd., 1923, No. 24. Quoted in Rev. Española de Med. Cirurg., March, vol. 7, No. 69, p. 163.

1897 INDIAN LANCET, Calcutta. Vol. 10, p. 146; vol. 10, p. 556, Kala-azar in Assam.
1885 INDIAN MEDICAL GAZETTE, Calcutta. Vol. 20, p. 83, "Kala-azar," or the black death of the Garo Hills.
1892 —— Vol. 27, pp. 49-51, Kala-azar and beri-beri in Assam.
1897 —— Vol. 32, p. 461, Kala-azar and diseases confused with it.
1897 —— Vol. 32, pp. 472-476.
1898 —— Vol. 33, p. 25, The kala-azar report and its critics.
1908 —— Vol. 43, pp. 221-222, Diagnosis and prognosis in kala-azar.
1909 —— March, vol. 44, pp. 105-106, The kala-azar case in Germany (Current Topics).
1909 —— May, vol. 44, pp. 190-191, Kala-azar and Leishman-Donovan bodies (Current Topics).
1910 —— December, vol. 45, p. 506, Parasitic granuloma (Current Topics).
1912 —— Kala-azar and bed-bugs. March. Vol. 47, No. 3, p. 108.
1912 —— Kala-azar and tropical sore. January. Vol. 47, No. 1, pp. 24-25.
1922 —— The problems of kala azar. June. Vol. 57, No. 6, pp. 221-224.
1897 INDIAN MEDICAL RECORD, Calcutta. Vol. 13, p. 364, Recent reports on kala-azar; is Rogers right?

1897 —— Vol. 13, pp. 404-406, The new theory of kala-azar: a plea for further scientific research.
1923 INDIAN MEDICAL RECORD, August and September, vol. 43, Nos. 8 and 9. (Kala-azar Numbers) I and II.
1910 INGRAM, Capt. A. C. Kala-azar. Annual Report of the Government General Hospital, Madras, for the year 1910.
1925 INNES, FLORA R. Three unusual cases of kala-azar. Journ. Assoc. Med. Women in India, February, vol. 13, No. 1, pp. 40-43.
1914 IOANNIDES, G. S. Kala-azar in Greece. (Ref. in Trop. Dis. Bull., April 15, 1915, p. 268.)
1917 IRUEGAS, F. Un caso de kala-azar infantil. Pediatria Españ., vol. 6, pp. 36-39.
1914 ISUPA, V. Azione di alcuni derivati della chinina sulla *Leishmania infantum* in vitro. Biochim. e Terap. Sper., Milano, 1913-1914, vol. 5, pp. 16-27.
1914 —— Azioni di alcuni derivati della chinina sulla *Leishmania infantum* in vitro. Boll. Accad. Gionenia Scienze Naturali in Catania, May, series 2, No. 31, pp. 60-62.
1923 IYENGAR, K. R. K. Studies in the value of the Wassermann test. No. V. Significance and value of a positive Wassermann reaction in kala-azar. Ind. Journ. Med. Res., July, vol. 11, No. 1, pp. 237-238.
1925 IZAR, G. Sur le traitement de la leishmaniose interne. March 7. Presse Méd., vol. 33, No. 19, p. 303.

1872 JACKSON, DR. Report on the Burdwan fever of Lower Bengal.
1916 JACKSON, T. A case of kala-azar treated by intravenous injection of tartar emetic at St. George's Hospital, Bombay. Ind. Med. Gazette, December, vol. 51, No. 12, p. 450.
1889 V. JAKSCH. Über Leukämie und Leukozytose in Kindesalter. Wiener klinische Wochenschr., vol. 2, pp. 435-437 and pp. 456-458.
1890 —— Über die Diagnose und Therapie der Erkrankungen des Blutes. Prager medizinische Wochenschr., vol. 15, pp. 389, 403, 414.
1905 JAMES, S. P. On kala-azar, malaria and malarial cachexia. Scientific memoirs by Officers of the Medical and Sanitary Departments of the Government of India. New series. No. 19.
1905 —— On kala-azar and malarial cachexia. The Lancet, December 23, p. 1845.
1913 JANNOT, A. Infection de la souris avec le virus de la leishmaniose canine naturelle. Bull. Soc. Path. Exot., December, vol. 6, No. 10, pp. 683-685.
1920 JAVARONE, N. Cronaca della leishmaniosi infantile a Napoli e dintorni. Pediatria, August, vol. 28, No. 15, pp. 716-723.
1901 JEANNACOPOULOS, C. Contribution à l'étude du ponos (Translation). Ann. Soc. Méd. de Gand., vol. 80, pp. 315 319.
1910 JEFFERYS, W. H., and MAXWELL, J. L. The diseases of China, including Formosa and Korea. 8 vols. London: John Bale, Sons and Danielsson, Ltd. (Kala-azar, pp. 130-134.)
1905 JEMMA, ROCCO. Cura delle anemie infantili. Il Policlinico, sez. pratica, vol. 12, Part 24, p. 750.
1900 —— Sobre un caso di anemia splenica infantile da corpi di Leishman osservata a Palermo. Il Policlinico, sez. pratica, vol. 16, Part 44, pp. 1381-1384.
1910 —— Sull' anemia splenica infantile da parassiti di Leishman (Kala-azar?). Riforma medica, Palermo, vol. 26, No. 13, pp. 319-323. Gazzetta di Medicina e Chir., Palermo, vol. 9, pp. 117-120.
1910 —— Über infantile Milzanämie durch Leishman'sche Parasiten (Kala-azar?). Deutsch. Arch. für klin. Med., vol. 100, Parts 5-6, pp. 466-486.
1910 —— Sulla infezione spontanea da parassita di Leishman nei cani. Pathologica, June, vol. 2, p. 250.
1912 —— Sulla leishmaniosi del cane nei dintorni di Palermo. Pathologica, August 1, vol. 4, No. 90, pp. 466-467.
1912 —— Considerazioni sopra sessantatré casi di anemia da leishmania osservati nella clinica pediatrica di Palermo. Riforma Medica, August 24, vol. 28, No. 34, pp. 925-930.
1912 —— Communication personnelle. (Leishmanioses.) Arch. Inst. Pasteur, Tunis. No. 3, p. 196.
1912 —— Leishmansche Anämie. Monatsschrift f. Kinderheilkunde, vol. 11, No. 7, Abt. Orig. 3, pp. 321-356.
1913 —— Anemia da leishmania. La Pediatria, January 31 (veröffentlicht, March 15), vol. 21, No. 1 (second series), vol 11), pp. 1-43.
1913 —— L'Anémie par leishmania. Arch. de Méd. des Enfants, October, vol. 16, No. 10, pp. 721-766.

1914 JEMMA, R. Brevi considerazioni su 110 casi di leishmaniosi inf. osservati nella clinica pediatrica di Pal. Pediatria, February, vol. 22, No. 2, pp. 81-94.
1914 —— Kurze Betrachtungen über 110 in der Kinderklinik zu Palermo beobachtete Fälle von kindlicher Leishmaniosis. Monatsschr. f. Kinderheilkunde, Orig., vol. 12, No. 11, pp. 659-672.
1916 —— La cura specifica della leishmaniosi nei bambini. Pediatria, January, vol. 2, No. 1, pp. 1-5.
1917 —— Courte notice historique sur l'emploi de l'antimoine dans le traitement de la leishmaniose interne. Bull. Soc. Path. Exot., October, vol. 10, No. 8, pp. 762-769.
1917 —— Breva nota storica sull' introduzione dell' antimonio nella terapia della leishmaniosi interna. Mal. et Malat. d. Paesi Caldi, September-December, vol. 8, Nos. 5-6, pp. 178-183.
1918 —— Brief historical notes on the introduction of antimony in the therapeutics of internal leishmaniosis. Journ. Trop. Med. and Hyg., January 1, vol. 21, No. 1, pp. 1-3.
1922 —— Cultura dei parassiti di Leishman dal sangue periferico e coltivabilita do essi in terreni con sangue umano. Pathologica, February 1, vol. 13, No. 317, pp. 71-73.
1923 —— Considerazioni sulla diagnosi e sulla terapia della leishmaniosi infantile. Pediatria, June 15, vol. 31, pp. 633-640.
1923 —— Leishmaniosis infantum. Ergebnisse der inneren Med. und Kinderheilkunde, vol. 23, pp. 505-647.
1924 —— A propos du kala-azar infantile dans la région Mediterranéenne. Paris Méd., vol. 14, pp. 155-159.
1924 —— Considerations on the diagnosis and therapy of leishmaniasis. Journ. Orient. Med., August, vol. 2, No. 3, pp. 228-232, with 1 plate.
1911 —— and DI CRISTINA, G. Anemia da leishmania nei bambini. Vol. 7, Congresso Pediatrico Italiano, Palermo, April 20-23. Relazione. Pathologica, June 1, vol. 3, No. 62, pp. 268-270.
1911 —— —— Über die Leishmania-Anämie der Kinder. Zentralbl. f. Bakt., Abt. Orig., June 24, vol. 59, Part 2, pp. 109-177.
1911 —— —— Sull' anæmia da leishmania nei bambini. Gazetta Medica Italiana. August 17, vol. 62, No. 33, pp. 324-327.
1910 —— —— and CANNATA, S. Infezione sperimentale da *Leishmania infantum* nei cani. La Pediatria, 2 series, vol. 8, No. 4, pp. 241-252.
1910 —— —— —— Experimentale Infektion mit Leishman infantum bei Hunden. Zentralbl. f. Bakt., Orig., vol. 57, Part 1, pp. 59-68.
1910 JERUSALEMY. Kala-azar infantile en Chine. Revue de Médecine et d'Hygiène Tropicales, vol. 7, Part 2, pp. 121-123.
1927 JESSNER, M. Wie sind die Aussichten einer Immunisierung gegen Hautleishmaniose und einer Therapie der Erkrankung mit Leishmaniavakzine? Arch. f. Schiffs- u. Trop.-Hyg., vol. 31, pp. 72-74.
1925 —— and AMSTER, SIEGFRIED. Leishmania vakzine—Leishmaniin-reaktion. Deut. med. Woch., May 8, vol. 51, No. 19, p. 784.
1918 JOHNSTONE, E. M. A study of the blood changes in kala-azar after splenectomy. Chin. Med. Journ., November, vol. 32, No. 6, pp. 505-513.
1904 JOURNAL OF THE ROYAL ARMY MEDICAL CORPS. Vol. 2, pp. 207-209. Dum-dum fever (Editorial).
1907 JOURNAL OF TROPICAL MEDICINE AND HYGIENE. Vol. 10, pp. 161-162. Professor Leonard Rogers' Milroy Lectures on kala-azar.
191 Vol. 14, p. 9. Is ponos kala-azar? Professor Galle's discovery.
191 —— Vol. 14, p. 13. Diagnosis of kala-azar.
191 —— Vol. 14, p. 30. Leishman-Donovan bodies in "ponos."
191 Leishmania and mosquitoes. December 1, vol. 14, No. 23, p. 363.
191 JOURNAL OF THE AMERICAN MEDICAL ASSOCIATION. Infantile splenic anæmia and kala-azar (Editorial). August 12, vol. 57, No. 7, pp. 566-567.
1926 JUNKIN, C. I. Kala-azar. A case occurring in New York. Journ. Amer. Med. Assoc., vol. 86, pp. 479-482.

1911 KALA-AZAR BULLETIN, No. I: Experimental kala-azar in animals, pp. 1-62.
1912 —— No. II: Infantile kala-azar (Nicolle), pp. 67-118.
1912 —— No. III: Kala-azar and oriental sore in India, pp. 123-170.
1913 Kala-azar Committee Report on the operations of the Kala-azar Committee. Proc. 3rd Meeting of the Gen. Mal. Comm. held at Madras, November 18, 19 and 20, 1912, pp. 34-35.
1926 KALA-AZAR COMMISSION. Report No. 1 (1924-1925). Indian Medical Research Memoir, February, Supplementary Series to the Indian Journal of Medical Research.

1925 KAPUR, N. C. Observations on the treatment of kala-azar with urea stibamine in the Medical Out-patient Department of the Medical College Hospital, Calcutta. Ind. Med. Gazette, May, vol. 60, pp. 206-210.

1925 KASSIRSKY, J. A., MOSHEIKO, W. M., and FEDULOW, A. W. Über Leishmaniose bei Erwachsenen in Turkestan. Arch. f. Schiffs- u. Trop.-Hyg., August, vol. 29, pp. 368-379. (Abstract.)

1923 KASUGA, K. A new culture medium for *Leishmania donovani*. Far Eastern Assoc. Trop. Med. Trans. Fifth Biennial Congress, Singapore, pp. 698-699.

1925 —— and TAMURA, I. [Cultivations of *Leishmania donovani* and some experiments on the immunization.] Gun-I-Dan Zasshi (J. Japan Milit. Med. Assoc.), No. 141. (Summarized in Japan Med. World, 1925, p. 295.)

1914 KATSAS, G. G. Case of kala-azar with discovery of parasites in the peripheral blood. Abstract in Trop. Dis. Bull., 1915, April 15, p. 260.

1918 KENNEDY, J. C. Six cases of kala-azar: (1) Their treatment; (2) Notes on the epidemiology. Journ. Roy. Army Med. Corps, Feb., vol. 30, No. 2, pp. 209-215.

1918 KERR, H. I. W. Kala-azar in Malta. Two cases treated by intravenous injection of tartar emetic. Lancet, July 13, pp. 45-46.

1916 KHARINA-MARINUCCI, R. Influenza dell' antimonio sulla curva febbrile nella leishmaniosi interna. Pediatria, vol. 24, December, No. 12, pp. 717-737.

1925 KLIGLER, I. J. The cultural and serological relationship of leishmania. Trans. Roy. Soc. Trop. Med. and Hyg., vol. 19, pp. 330-335.

1921 KLIPPEL and MONIER-VINARD. Un cas de kala-azar d'origine marocaine. Bull. et Mém. Méd. Hôp. de Paris, July 14, No. 24, pp. 1037-1038.

1922 —— Premier cas de kala-azar d'origine marocaine. Guérison par l'acetyl-aminophenyl stibinate de soude (stibenyl). Bull. et Mém. Soc. Méd. Hôp. de Paris, Jan. 10, year 38, 3rd series, No. 2, pp. 76-85.

1918 KNOWLES, R. Notes on some results in kala-azar. Ind. Journ. Med. Res., April, vol. 5, No. 4, pp. 528-566.

1918 / 1920 —— King Edward VII Memorial Pasteur Institute. The First, Second and Third Annual Reports for 1917, 1918 and 1919. Assam Secretariat Printing Office.

1920 —— A study of kala-azar: Parts I to IV. Ind. Journ. Med. Res., July, vol. 8, No. 1, pp. 140-209.

1923 —— Kala-azar. Lecture Notes in Medical Protozoology, pp. 60-77.

1924 —— The Indian Medical Year, 1923, a Review. The Indian Medical Gazette.

1925 —— The Indian Medical Year, 1924, a Review. The Indian Medical Gazette.

1926 —— The Indian Medical Year, 1925, a Review. The Indian Medical Gazette.

1927 —— The Indian Medical Year, 1926, a Review. The Indian Medical Gazette.

1927 —— Medical Protozoology (in the Press).

1924 —— and DAS GUPTA, B. M. The diagnosis of kala-azar by examination of thick blood films. Ind. Med. Gazette, September, pp. 438-440.

1924 —— —— —— On transient infections with *Leishmania donovani* in men and animals. Ind. Med. Gaz., June, vol. 59, No. 6, pp. 292-295.

1923 —— NAPIER, L. E., and DAS GUPTA, B. M. Kala-azar transmission problem. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 321-349.

1924 —— —— and SMITH, R. O. A. A preliminary note on a herpetomonas found in the gut of the sand-fly, *Phlebotomus argentipes*, fed on kala-azar patients. Ind. Med. Gazette, December, vol. 59, No. 12, pp. 593-597.

1925 —— —— —— —— The vector of kala-azar. Notes in Brit. Med. Journ., Jan. 17, 1925, pp. 132, and China Med. Journ., March, vol. 39, No. 3, p. 270.

1921 KNUTH, DR. PHIL P., and DR. PHIL ET MED. VET. P. J. DU TOIT. Die Leishmaniose. Handbuch der Tropenkrankheiten, herausgegeben von Prof. Dr. Carl Mense, Kassel, Zweite auflage, Sechster.

1914 KOHL-YAKIMOFF, N. K., YAKIMOFF, W. L., and SCHOKHOR, N. H. The question of leishmaniasis in Russia. (1) Leishmaniasis in the dog in Tashkent. (2) Leishmaniasis in children. (3) Leishmaniasis in adults. (4) A case of oriental sore in a dog. Russky Vrach, Nos. 8 and 9.

1915 KOKORIS, D. Über die Splenektomie bei Kala-azar. Münch. med. Woch., July 27, No. 30, vol. 62, pp. 1008-1009.

1913 KORKE, V. T. Progress report on "Some observations on the epidemiology of kala-azar in Madras." Proc. 3rd Meet. Gen. Mal. Com. (Madras), 1912, November 18-20, pp. 230-256.

1914 —— A note on the production of localised lesions by leishmania donovani in *Macacus sinicus*. Ind. Journ. Med. Res., April, vol. 1, No. 4, pp. 622-625.

1918 KORNS, J. H. Antimony in kala-azar. China Med. Journ., January, vol. 32, No. 1, pp. 26-29.

1926 KORCHITZ, E. V. On the treatment of infantile leishmaniasis with stibenyl. Pensée Médicale d'Uzbekistane Tashkent, vol. 1 (V), No. 1, pp. 36-37. (In Russian.)

1900 KRONECKER, F. Die Kala-azar in der vorderindischen Provinz Assam; eine tropen-pathologische Studie nach englischen Quellen dargestellt. Archiv für Schiffs- und Tropen-Hygiene, vol. 4, pp. 220-238.
1919 KUNDU, S. S. Report on kala-azar treated in the Nowgong Earle Hospital since 1917. Ind. Med. Gazette, October, vol. 54, No. 10, pp. 376-379.
1920 —— A study of kala-azar. Ind. Journ. Med., October, vol. 1, No. 3, pp. 155-161.
1920 —— Further observations on kala azar. Ind. Med. Gazette, vol. 55, February. No. 2, pp. 53-57.
1925 —— Intramuscular injection of urea stibamine. Ind. Med. Gaz., December, vol. 60, No. 12, pp. 583-584.
1925 —— An antimony rash. Ind. Med. Gaz., May, p. 247. (Correspondence.)
1925 KUHN, PH. and SCHMIDT, HANS. Neuere Erfahrungen mit Antimonpräparaten bei Tropenkrankheiten. Vortr. auf d. Tag. der Deutsch. Tropenmed. Ges., Hamburg.

1923 LABBÉ, M. Le kala-azar infantile d'origine française. Bull. et Mém. Soc. Méd. Hôp. de Paris, January 18, 3rd series, year 39, No. 1, p. 10.
1918 —— TARGHETTA and AMEUILLE. Le kala-azar infantile en France. Bull. Acad. de Méd., April 2, 3rd series, vol. 79, year 82, No. 13, pp. 288-290.
1918 —— —— —— Le kala-azar infantile en France. Brit. Journ. Children's Diseases, April-June, vol. 15, Nos. 172-174, pp. 120-122, and Bull. Acad. de Méd., April 2, 3rd series, vol. 79, year 82, No. 13, pp. 280-290.
1913 LA CAVA, F. Un caso di leishmaniosi interna (kala-azar) in una giovinetta di 14 anni. Mal. e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 317-320.
1914 —— Un caso di leishmaniosi interna (kala-azar) in una giovinetta di 14 anni. Pathologica, March 15, vol. 6, No. 129, pp. 151-153.
1915 LAFONT, A., and HECKENROTH, F. Un cas de leishmaniose canine à Dakar. Bull. Soc. Path. Exot., April, vol. 8, No. 4, pp. 162-164.
1910 LAMFERT, ALEXANDER C. Kala-azar in the Yangtse Valley. Brit. Med. Journ., March 26, vol. 1, p. 750.
1898 LANCET. A case of kala-azar in Vienna Vol. 2, p. 1570.
1908 —— Cases of kala-azar. (Society of Tropical Medicine and Hygiene.) Vol. 1, p. 719.
1909 —— The parasite of kala-azar. (Annotation.) February 6, vol. 1, p. 415.
1909 —— Kala-azar in Sicily. (Report of the 16th International Medical Congress, August 24 to September 4.) September 18, p. 874.
1911 —— Kala-azar and the mosquito. (Annotation.) November 4, vol. 2, pp. 1285-86.
1912 —— The announcement of discovery by Capt. W. S. Patton, I.M.S., of development of the parasite of kala-azar in Indian and European bed-bugs. February 17, vol. 182, No. 4616, p. 447.
1912 —— Kala-azar and the bed bug. (Annotation). February 24, vol. 182, No. 4617, p. 520.
1917 —— Preparation of stable colloidal antimony by Dr. Brahmachari, February 24.
1903 LAVERAN, A. Présentations d'ouvrages, etc. (*Piroplasma donovani.*) Bull. de l'Académie de Médecine, séance of November 3, 3rd series, vol. 1, No. 35 pp. 239-240.
1904 —— Présentation de Parasite. (*Piroplasma donovani* en Tunisie d'après une observation de Cathoire.) Bull. de l'Académie de Médecine, March 22, 3rd series, vol. 51, pp. 247-248.
1909 —— Leishmanioses. La Presse Médicale, April 10, vol. 17, pp. 257-258.
1912 —— Infection généralisée de la souris par la *Leishmania donovani.* Comptes Rendus de l'Acad. des Sciences, February 26, vol. 154, No. 9, pp. 559-561.
1912 —— Infections des souris et des rats dues au kala-azar Méditerranéen et au kala-azar Indien. Bull. Soc. Path. Exot., November, vol. 5, No. 9, pp. 715-721.
1913 —— Les macaques et les chiens sont sensibles au kala-azar Indien comme au kala-azar Méditerranéen. C. R. Acad. Sciences, November 17, vol. 157, No. 20, pp. 898-901.
1913 —— Kala-azar Méditerranéen et kala-azar Indien. Bull. Soc. Path. Exot., October, vol. 6, No. 8, pp. 574-579.
1913 —— Au sujet de l'historique du kala-azar Méditerranéen. Bull. Soc. Path. Exot., January, vol. 6, No. 1, pp. 23-24.
1913 —— Infections du cobaye, du lapin et du chat par la *Leishmania infantum.* Bull. Soc. Path. Exot., February, vol. 6, No. 2, pp. 110-114.
1913 —— Présentation d'un chien attient de leishmaniose et du keratite. Bull. Soc. Path. Exot., vol. 6, No. 7, pp. 447-448.

1913 —— Les leishmanioses. (Abstract from report.) Trans. XVIIth Intern. Congress of Medicine, London. Section XXI. Trop. Med. and Hyg., Part 2, pp. 179-182.
1914 —— Au sujet d'un cas de leishmaniose canine signalé à Marseilles. Bull. Soc. Path. Exot., March, vol. 7, No. 3, pp. 173-174.
1914 —— Nouveaux faits tendant à démontrer que le kala-azar Méditerranéen doit être identifié au kala-azar Indien. C. R. Acad. Sciences, April 14, vol. 158, No. 15, pp. 1060-1064.
1914 —— Les leishmanioses chez les animaux. Ann. Inst. Pasteur, September-October, vol. 28, Nos. 9-10, pp. 823-838, and November-December, Nos. 11-12, pp. 885-912.
1915 —— Sur une culture de *Leishmania donovani* souillée par un champignon. Bull. Soc. Path. Exot., July, vol. 8, No. 7, p. 429
1915 —— Les leishmanioses chez les animaux. Ann. Inst. Pasteur, January, vol. 29, No. 1, pp. 1-21, and February, No. 2, pp. 71-104.
1915 —— Des lacertiens peuvent-ils être infectés par des leishmania? Bull. Soc. Path. Exot., March, vol. 8, No. 3, pp. 104-109.
1916 —— Au sujet de l'historique de la leishmaniose viscérale. Bull. Soc. Path. Exot., February, vol. 9, No. 2, pp. 74-75.
1917 —— Leishmanioses. Masson and Co., Paris.
1918 —— Sur les leishmanioses expérimentales, et un particulier sur la leishmaniose canine, chez le souris blanche. Bull. Soc. Path. Exot., March, vol. 2, No. 3, pp. 205-216.
1921 —— and FRANCHINI, G. Sur un herpetomonas du Loir. Bull. Soc. Path. Exot., vol. 14, No. 5, p. 278.
1917 —— and HAVET, J. Contribution à l'étude de la leishmaniose viscérale naturelle du chien. Bull. Soc. Path. Exot., May, vol. 10, No. 5, pp. 386-392.
1903 —— and MESNIL, F. Sur un protozoaire nouveau *Piroplasma donovani* (Laveran and Mesnil) parasite d'une fièvre de l'Inde. Comptes Rendus de l'Académie des Sciences, December 7, vol. 137, pp. 957-962.
1904 —— —— Nouvelles observations sur *Piroplasma donovani*, Laveran et Mesnil. Comptes Rendus de l'Académie des Sciences, January 25, vol. 138, pp. 187-189.
1904 —— —— Un protozoaire nouveau parasite d'une fievre de l'Inde. Annales d'Hygiène et de Médecine coloniales, vol. 7, No. 2, p. 226.
1904 —— —— On a new Protozoon (*Piroplasma donovani*, Laveran and Mesnil). The parasite of an Indian fever. (Translation from Comptes Rendus de l'Académie des Sciences, vol. 137, pp. 957-962.) Journ. Roy. Army Med. Corps, vol. 2, pp. 216-218.
1913 —— and NICOLLE, C. Le kala-azar Méditeranéen ou infantile. Transactions, XVIIth Inter-Congress of Medicine, London, Sec. XXI, Trop. Med. and Hyg., Part I, pp. 71-107.
1909 —— and PETTITT, A. Infections légères du rat et de la souris par la *Leishmania donovani*. C. R. Soc. Biol., Paris, July 5, vol. 116, pp. 911-913.
1909 —— —— Infections expérimentales légères ou latentes du singe et du chien par le kala-azar Tunisien. Bull. Soc. Path. Exot., Paris, December 8, vol. 2, No. 10, pp. 584-587.
1909 —— —— Infections légères du cobaye par *Leishmania donovani*. Comptes Rendus de la Société de Biologie, July, 3, vol. 67, p. 8.
1910 —— —— Sur les cultures de *Leishmania donovani* en milieu liquide. Bull. Soc. Path. Exot., April 13, vol. 3, No. 4, pp. 216-217.
1910 —— —— Culture de la *Leishmania donovani* en milieu liquide. Comptes Rendus de la Société de Biologie, 22 Janvier, vol. 68, p. 114. (Erratum, p. 276.)
1922 LEAO, A. E. DE A. A reacçao de Wassermann na leishmaniose. Mem. Ins. Oswaldo Cruz., vol. 15, No. 1, pp. 209-219.
1911 LEBOEUF. Le kala-azar Indien et le kala-azar infantile, pp. 309-335. Paraluidisme et fièvres des pays chauds. (Traité de Pathologie Exotique.) P. 378, fig. 26, vol. 8. Paris: Librairie J. B. Baillière et Fils, 19, rue Hauteville.
1910 LEDINGHAM, J. C. G. Kala-azar in Mesopotamia. Brit. Med. Journ., July 19, p. 88.
1926 LEGER, MARCEL. Relationship of the leishmanias. Rev. Prat. Malad. des Pays Chauds, February, year 4, vol. 5, No. 10, pp. 439-445.
1921 LEGROUX, RENÉ, and JIMENEZ, J. Facteur des croissances dans les cultures de *Leishmania donovani*. C. R. Acad. Sci., December 19, vol. 173, No. 25, pp. 1423-1425.
1903 LEISHMAN, W. B. On the possibility of the occurrence of trypanosomiasis in India. Brit. Med. Journ., May 30, pp. 1252-1254, and November 21, pp. 1376-1377.
1904 —— Note on the nature of the parasites found in tropical splenomegaly. Brit. Med. Journ., February 6, vol. 1, p. 303.

1904 LEISHMAN, W. B. The nature of the Leishman-Donovan bodies. Brit. Med. Journ., July 2, vol. 2, p. 29.
1904 —— On the Leishman-Donovan body. Brit. Med. Journ., September 17, vol. 2, pp. 642-645.
1904 —— The nature and significance of the Leishman-Donovan body. Journ. Trop. Med. and Hyg., vol. 7, pp. 258-259.
1904 —— Notes upon the further investigation of the parasites of kala-azar and Delhi boil. Journ. Roy. Army Med. Corps, vol. 3, pp. 287-292.
1904 —— Dum-Dum fever? Kala-azar? Non-malarial remittent fever? Journ Roy. Army Med. Corps, vol. 2, p. 623.
1906 —— Kala-azar oder Innere Leishmaniasis. Handbuch der Tropenkranheiten, edited by Prof. Dr. Carl Mense, 2 auflage, band 4, Ambrosius Barth, Leipzig.
1907 —— Kala-azar. A system of medicine edited by various writers, edited by T. C. Clifford Allbutt and H. D. Rolleston, vol. 2, part 2, Macmillan and Co.
1911 —— Critical review. Kala-azar and tropical sore. Quar. Journ. of Med., October, vol. 5, No. 17, pp. 109-152. Reprinted in the Journ. Roy. Army Med. Corps, December, vol. 17, No. 6, pp. 567-580.
1912 —— A critical review of kala-azar and tropical sore. Journ. Roy. Army Med. Corps, January, vol. 18, No. 1, pp. 1-20.
1912 —— A critical review of kala-azar and tropical sore. Journ. Roy. Army Med. Corps, February, vol. 18, No. 2, pp. 125-137.
1905 —— and STATHAM, J. C. B. The development of the Leishman body in cultivation. Journ. Roy. Army Med. Corps, March, vol. 4, pp. 321-333.
1922 LEIVA, L. The cultivation of *Leishmania infantum* and *Leptomonas ctenocephali* on Triple N. medium. Philippine Journ. Sci., vol. 20, No. 2, pp. 179-183.
1911 LEMAIRE, G. Algérian case of infantile kala azar. Bull. Soc. Path. Exot., Paris, vol. 4, No. 6, p. 346.
1911 —— Premier cas de leishmaniose algérienne. Bul. Soc. Path. Exot., Oct. 11, vol. 4, No. 8, pp. 554-563.
1914 SERGENT, ED., and L'HERITIER, A. La leishmaniose naturelle du chien à Alger. Étude clinique et anatomo-pathologique. Rev. Med. d'Alger., January, pp. 1-14.
1914 —— —— —— Spécificité de la kératite observée chez les chiens atteints de leishmaniose naturelle. Bull. Soc. Path. Exot., March, vol. 7, No. 3, pp. 193-196.
1913 —— —— —— Recherches sur la leishmaniose du chien d'Alger. Bull. Soc. Path. Exot., October, vol. 6, No. 8, pp. 570-581.
1925 LEMIERRE, A., LEON-KINDBERG, M., and BERNARD, F. Kala azar Tunisien à évolution mortelle chez une adulte. Bull. et Mém. Soc. Méd. d'Hôp., 3e s. vol. 40, pp. 537-546.
1926 LEPINE, PIERRE. Three cases of infantile kala-azar from Syria. Bull. Soc. Path. Exot., June 9, vol. 19, No. 6, pp. 429-431.
1920 LESLIE, PERCY E. Kala-azar. Canadian Med. Assoc. Journ., August, vol. 10, No. 8, pp. 724-728.
1912 LEVY, A. La réaction meiostagminique d'ascoli dans le kala-azar expérimental du chien, dans l'infection par le bacille de Friedländer, le staphylocoque pyogène doré et le bacille de la tuberculose chez le lapin et le cobaye. Arch. Inst. Pasteur Tunis, Part 2, pp. 75-95.
1909 LEVY, E., and LEVY, A. Un cas de pseudo-leucémie ou lymphadénie splénique. Bull. Soc. Méd. Tunis, October 27, 8me année, Part 1, pp. 13-17.
1909 —— and ORTONA, C. Un cas de kala-azar infantile. Bull. Soc. Méd. Tunis, October 27, 8me année, Part 1, pp. 12-18.
1924 LEVY, P. P. Sur l'hématologie du kala-azar infantile (à propos d'un cas observé a Paris et terminé par la guérison). Bull. Soc. Path. Exot., June 11, vol. 17, No. 6, pp. 477-482, with 2 charts in text.
1910 LICCIARDI, S. Contributo clinico allo studio del kala-azar in Catania. Gazzetta Medica di Roma, vol. 36, pp. 197-205. (Malpighi, April.)
1911 LIGNOS, A. Quinze cas de kala-azar infantile observés à Hydra. Bull. Soc. Path. Exot., séance of December 13, vol. 4, No. 10, pp. 664-666.
1912 —— Un cas de kala-azar infantile se terminant par le guérison. Bull. Soc. Path. Exot., séance of February 14, vol. 5, No. 2, pp. 91-93.
1912 —— Absence des leishmania à l'autopsie d'un enfant mort de kala-azar. Bull. Soc. Path. Exot., June, vol. 5, No. 6, pp. 349-351.
1912 —— Déplacement de la rate chez un enfant atteint de kala-azar. Bull. Soc. Path. Exot., séance of May 8, vol. 5, No. 5, pp. 271-273.
1912 —— La gangrène de la bouche et du nez dans le kala-azar infantile d'Hydra. Bull. Soc. Path. Exot., séance du May 8, vol. 5, No. 5, pp. 270-271.
1913 —— L'Infection par leishmania des chiens de l'Ile d'Hydra. Bull. Soc. Path, Exot., February, vol. 6, No. 2, p. 117.

1913 —— Deuxième cas de guérison de kala-azar infantile observé à Hydra. Bull. Soc. Path. Exot., June, vol. 6, No. 6, pp. 430-432.
1913 —— Un cas de fièvre réellement noire (kala-azar) observé à Hydra. Bull. Soc. Path. Exot., February, vol. 6, No. 2, pp. 114-117.
1914 —— Troisième cas de guérison de kala-azar infantile observé à Hydra. Bull. Soc. Path. Exot., January, vol. 7, No. 1, pp. 43-45.
1914 —— La mortalité par kala-azar à Hydra pendant l'année 1911. Bull. Soc. Path. Exot., March, vol. 7, No. 3, p. 193.
1914 —— De l'époque de l'apparition du kala-azar à Hydra. Bull. Soc. Path. Exot., January, vol. 7, No. 1, pp. 45-46.
1915 —— Quelques nouveaux cas de guérison de kala-azar infantile observés à Hydra. Bull. Soc. Path. Exot., January, vol. 8, No. 1, pp. 25-28.
1916 —— La leishmaniose canine à Hydra. Bull. Soc. Path. Exot., May, vol. 9, No. 5, p. 302.
1925 LING, W. P. Further observations on the fundus oculi of kala-azar patients. China Med. Journ., August, vol. 39, pp. 681-683.
1925 LLOYD, R. B., NAPIER, L. E., and SMITH, R. O. A. The blood meal of *Phlebotomus argentipes* identified by precipitin antisera. Ind. Journ. Med. Res., vol. 12, No. 4, pp. 811-817.
1913 LOMBARDO, G. Contributo allo studio delle alterzione anatomiche dell' anemia da leishmania. Pathologica, May 15, vol. 5, No. 109, pp. 292-296.
1804 LONGO, A. Contributo al valore diagnostico delle cellule eosinofile nelle varie forme di anemia dei bambini: ricerche cliniche e microscopiche. La Pediatria, vol. 2, pp. 72-86.
1910 —— Sulla coltivabilità della *Leishmania infantum* nel sangue splenico infetto citratato. Policlinico (sez. pratica), May, vol. 17, Part 21, pp. 643-644.
1910 —— Sopra un caso di anemia splenica a leishmania a decorso acuto. Rivista di Clinica Pediatrica, vol. 8, Part 7, pp. 597-604.
1912 —— Tentativi immuno-diagnostici ed immunoterapeutici nella leishmaniosi infantile. Policlinico (sez medica), October, vol. 19, No. 10, pp. 446-452.
1917 —— Sopra alcuni casi di kala-azar infantile trattati col tartaro stibiato. Pediatria, August, vol. 25, No. 8, pp. 449-468.
1890 DI LORENZO, G. Contribuzione alla casuistica e clinica dell' anemia splenica infettiva dei bambini. Arch. Italiano di Pediatria, vol. 8, pp. 175-188.
1913 LO RE, M., and DE STEFANO, S. Sopra otto casi di anemia da leishmania. Gaz. Internaz. d. Med. Chirurg. Igiene, December 6, No. 49, pp. 1157-1161.
1904 LOW, G. C. Discussion on the Leishman-Donovan body. Brit. Med. Journ., vol. 2, p. 658.
1910 —— Kala-azar in Mesopotamia. Brit. Med. Journ., July 26, pp. 115-116.
1910 —— Kala-azar in Mesopotamia and its incubation period. Brit. Med. Journ., December 6, pp. 758-759.
1910 —— Intravenous injections of antimonium tartaratum in kala-azar. Brit. Med. Journ., June 7, pp. 702-704.
1926 —— and COOKE, W. E. A case of congenital kala-azar. Lancet, vol. 2, pp. 1209-1211.
1927 —— and SAYERS, E. G. Early diagnosis of kala-azar. Journ. Trop. Med., Feb. 15, vol. 30, pp. 46-68.
1910 LUENGO, PABLO, DE BUEN, SADI, and LUENGO, E. Siete casos de kala-azar infantil. Arch. de Cardiologia y Hematologia, Madrid, October, No. 10, pp. 369-383.
1911 LUNA, FRANCESCO. XXV. caso di anemia da leishmania osservato a Palermo La Pediatria, January, series 2, vol. 9, No. 1, pp. 45-47.
1914 —— Particolarità culturali del parassita di Leishman nel terreno di N N N. Pathologica, September 1, vol. 6, No. 140, pp. 443-445.
1914 —— Culture della *Leishmania donovani* in aerobiosi e in anaerobiosi nel terreno Novy-Neal-Nicolle. Lavori d. Soc. Italiana di Patologia Esotica, pp. 57-60.
1925 LWOFF, ANDRE. Croissance hypertrophique devision multiple et constitution de la cinetide de *Leishmania donovani* Lav. et Mes. type infantum, Ch. N. C. R. Soc. Biol., Jan. 30, vol. 92, No. 3, pp. 160-163, with 20 figs.

1921 MCDONAGH, J. E. R. Some remarks on the development of the Leishman-Donovan bodies. Brit. Journ. Derm. and Syph., May, vol. 33, No. 5 (No. 391), pp. 182-188; Journ. Trop. Med. and Hyg., March 15, vol. 24, No. 6, pp. 77-79; and Proc. Roy. Soc. Med. (Sect. of Dermat.), April, vol. 14, No. 6, pp. 55-58.
1904 MCFARLAND, J. The Leishman-Donovan blood parasites. Amer. Med., vol. 7, No. 23, pp. 888-891.
1904 MCKENZIE, J. Enlargement of the spleen in Lower Bengal. Journ. Roy. Army Med. Corps, vol. 3, pp. 356-359.

1905 McKENZIE, J. A case of Dum-Dum fever (kala-azar). Journ. Roy. Army Med. Corps, vol. 5, pp. 628-630.
1906 —— Kala-azar. Journ. of the Roy. Army Med. Corps, vol. 6, pp. 163-164.
1905 MACKIE, F. P. Leishman-Donovan disease. The Lancet, vol. 2, p. 185.
1907 —— Note on an unsuccessful attempt to convey kala-azar to animals. Brit. Med. Journ., vol. 1, p. 1363.
1913 —— Progress report on kala-azar. Proceedings of the Third Meeting of the General Malaria Committee, Madras, November 18, 19, and 20, 1912, p. 233-238.
1914 —— Note on some bodies of unknown nature found in fæces of kala-azar patients. Ind. Journ. Med Res., October, vol. 2, No. 2, pp. 510-515.
1914 —— The progress of kala-azar in a localized community. Ind. Journ. Med. Res., October, vol. 2, No. 2, pp. 505-509.
1914 —— Kala-azar in Nowgong (Assam). Ind. Journ. Med. Res., April, vol. 1, No. 4, pp. 626-662.
1914 —— Kala-azar in Assam. Precis of a Progress Report, February to September, 1913. Proc. Third All-India Sanitary Conference, Lucknow. Supplement. Ind. Journ. Med. Res., vol. 5, pp. 12-14.
1914 —— A flagellate infection of sand-flies. Ind. Journ. Med. Res., vol. 2, No. 1, p. 377.
1915 —— The presence of leishmania in the peripheral blood of cases of kala-azar in Assam. Ind. Journ. Med. Res., July, vol. 3, No. 1, pp. 90-92.
1915 —— Tartar emetic in kala-azar. Brit. Med. Journ., November 20, p. 745.
1915 —— Insects and kala-azar. Ind. Journ. Med. Res., April, vol. 2, No. 4, pp. 942-949.
1915 —— The experimental transmission of Indian kala-azar to animals. Ind. Journ. Med. Res., April, vol. 2, No. 4, pp. 934-941.
1921 —— Stibenyl. (Correspondence.) Ind. Med. Gazette, February, vol. 56, No. 2, pp. 76-77.
1922 —— The problem of kala-azar. Ind. Med. Gazette, September, vol. 57, No. 9, pp. 326-331.
1923 —— DAS GUPTA, B. M., and SWAMINATH, C. S. Progress report on kala-azar (work carried out in Shillong between June and November, 1921). Ind. Journ. Med. Res., October, vol. 2, No. 2, pp. 501-509.
1923 —— and PATNI, H. C. The evidence of cure in the treatment of kala azar by antimony. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 293-295.
1904 MACLEOD, KENNETH. Discussion on the Leishman-Donovan body. Brit. Med. Journ., vol. 2, p. 658.
1912 MACOTTA, G. Vingt-septième observation Tunisienne de kala-azar. Arch. l'Inst. Pasteur de Tunis, Part 2, pp. 61-62.
1907 MADRAS GOVERNMENT GENERAL HOSPITAL. Annual Report for 1907. Madras Government Press. (Abstracted in Ind. Med. Gazette, 1908, vol. 43, pp. 230-231 and 233-234.)
1908 —— Annual Report for 1908. Madras Government Press.
1914 MAGAUDDA, P. Sulle complicanze nervose nella leishmaniosi interna. Lav. d. Soc. Italiana di Patologia Esotica, pp. 107-110.
1914 MAGGIORE, S. Contemporanea infezione di corpi Leishman e di micrococco di Bruce in un bambino di 10 mesi. Lav. d. Soc. Italiana di Patologia Esotica, pp. 62-63.
1916 —— Contributo allo studio della patogenesi dell' anemia nella leishmaniosi interna. Mal. e Malat. d. Paesi Caldi, January, vol. 7, No. 1, pp. 18-20.
1925 —— Contributo allo studio etio. Patogenetico della infezione da leishmania. Pediatria, Feb. 15, vol. 33, No. 4, pp. 169-172.
1917 —— and SINDONI, M. Sulla presenza di leucotossine circolanti nel siero di sangue di infermi di leishmaniosi interna. Pediatria, February, vol. 25, No. 2, pp. 81-88.
1924 MAITRA, J. N. A flagellate found in the peripheral blood-smear of a case clinically one of kala-azar. Calcutta Med. Journ., February, vol. 18, No. 8, pp. 618-619.
1925 MAITRA, JODENDRA N. Is kala-azar contagious? Cal. Med. Journ., 1925-26, vol. 20, pp. 315-317.
1925 MAJUMDER, B. L. A report on 202 cases of kala-azar treated in Campbell Medical Hospital in 1923. Cal. Med. Journ., July, pp. 10-13.
1911 MAKKAS, G. N. L'ablation totale de la rate comme moyen thérapeutique du "Ponos." Communication faite à la Société Médicale d'Athènes. Séances March 5 and May 28.
1911 —— and PAPASSOTIRION. Nouveau procédé diagnostic du "Ponos." Communication faite à la Société Médicale d'Athènes Séance May 28,

1921 MALLARDI, MARIO. Il ricambio dell antimonio nella leishmaniosi interna. Pediatria, October 1, vol. 29, No. 19, pp. 878-885
1921 —— Degenerazione cistica della milza nel corso della leishmaniosi interna. Pediatria, October 15, vol. 29, No. 20, pp. 934-937.
1922 —— Contributo statistico-clinico alla terapia specifica della leishmaniosi interna. Pediatria, October 1, vol. 30, No. 19, pp. 889-897.
1923 Comportamento del sangue e degli organi emopoietici nella leishmaniosi sotto l'influenza della terapia. Pediatria, vol. 31, March 1, No. 5, pp. 230-246.
1924 —— I primi due casi di leishmaniosi curati a Bari. Pediatria, January 1, vol. 32, No. 1, pp. 47-49.
1925 —— Ancora sulla leishmaniosi infantile in provincia di Bari. Pediatria, April 1, vol. 33, No. 7, pp. 383-384.
1911 MANCEAUX, L. Sur la technique de culture des leishmania. Bull. de la Soc. de Path. Exot., 10 Mai, vol. 4, No. 5, pp. 280-288.
1921 MANSON-BAHR, PHILIP. Intravenous injection of stibenyl in kala-azar. Lancet, May 7, p. 991.
1921 —— Intravenous injection of stibenyl in kala-azar. Lancet, July 24, pp. 178-180.
1925 —— The technique of splenic puncture in the diagnosis of kala-azar. Lancet, July, vol. 2, pp. 70-71.
1925 —— Manson's Tropical Disease, 8th edition, revised and enlarged.
1904 MANSON, SIR PATRICK. Discussion on the Leishman-Donovan body. Brit. Med. Journ., London, vol. 2, pp. 657-658.
1905 —— Notes on two cases of febrile tropical splenomegaly (kala-azar) and a suggestion. Brit. Med. Journ., vol. 2, pp. 1261 1263. Lancet, vol. 169, p. 386. Journ. Trop. Med. and Hyg., vol. 8, p. 247.
1908 —— My experience of trypanosomiasis in Europeans and its treatment by atoxyl and other drugs. Ann. Trop. Med. and Parasit., vol. 2, No. 1, pp. 33-51. (Case of kala azar, p. 50.)
1908 —— A case of kala-azar: Recovery. (With discussion: Sir R. Havelock Charles, Major Leishman, Dr. Low, Dr. Harford, Dr. Sambon, D. Sandwith, and Fleet Surgeon Bassett-Smith.) Trans. Soc. Trop. Med. and Hyg., 1907-8, vol. 1, pp. 126-144.
1908 —— A case of kala-azar: Recovery. Journ. Trop. Med and Hyg., vol. 11, pp. 86-91.
1909 —— Demonstration of a case of kala-azar, apparently cured. Trans. Soc. Trop. Med. and Hyg., 1908-9, vol. 2, No. 5, pp. 169-171. Brit. Med. Journ., April 3, vol. 1, p. 843. Journ. Trop Med. and Hyg., June 1, vol. 12, No. 11, p. 167.
1909 —— The significance of fever in patients from the tropics. Medical Review, vol. 12, pp. 184-188. (Kala-azar, pp. 187-188.)
1904 —— and LOW, G. C. The Leishman-Donovan body and tropical splenomegaly. Brit. Med. Journ., vol. 1, pp. 183 186.
1904 —— —— The Leishman-Donovan body. Brit. Med. Journ., vol. 1, p. 1252.
1912 MANTOVANI, M. Infezione sperimentale da *Leishmania donovani* nel Conoglio. Nota preventiva. Pathologica, July 15, vol. 4, No. 89, p. 415.
1904 MARCHAND. Über neue Protozoeninfektionen beim Menschen. Münchener medizinische Wochenschrift, vol. 51, p. 630.
1904 —— and LEDINGHAM, J. C. G. Zur Frage der Trypanosoma-Infektion beim Menschen. Zentralblatt für Bakteriologie, 1 Abt. Orig., vol. 35, No. 5, pp. 594-598.
1904 —— —— Über Infektion mit "Leishman'schen Korporchen" (Kala-azar?) und ihr Verhaltnis zur Trypanosomenkrankheit. Zeitschrift für Hygiene und Infektions Krankheiten, vol. 47, pp. 1 40, mit 2 Tafeln.
1904 —— —— On the question of trypanosoma infection in man. Lancet, January 16, vol. 1, pp. 149-150.
1908 MARSHALL, D. G., and GULLAND, G. L. A case of kala-azar. Trans. Medico-Chirurg. Soc. Edin., 1907-8. New series, vol. 27, pp. 186-194.
1908 —— —— A case of kala-azar. Lancet, vol. 2, pp. 443-446.
1911 MARSHALL, W. E. Experimental kala-azar in the grey monkey of the Sudan (*Cercopithecus sabaeus*). Journ. Roy. Army Med. Corps, September, vol. 17, No. 3, pp. 255-262.
1911 —— Report of the Kala-azar Commission to investigate the prevalence and cause of the disease in the Eastern Sudan. Pathological report. Fourth report of the Wellcome Research Laboratory, vol. A., p. 157.
1912 —— Further experimental investigation into Sudan kala-azar. Journ. Roy. Army Med. Corps, September, vol. 19, No. 3, pp. 276-280.
1914 MARTINEZ, F. F. Quelques donnés sur le kala-azar infantil, 1914-15. Arch. de Mal. de l'Appar. Digest (etc.), vol. 8, pp. 600-603.

1915 MARTINEZ, F. F. El kala-azar infantil en la peninsula Iberica. Arch. Brasil. de Méd., November, vol. 5, No. 11, pp. 392-422.
1919 —— Treatment of leishmaniosis. Med. Ibera, Madrid, May 3, vol. 7, No. 78, p. 104. (Summarized in Journ. Amer. Med. Assoc., July 19.)
1926 —— Pathogenic leishmaniasis. Prensa Méd. Argent., vol. 13, pp. 426, 468, 605.
1916 MARTINI, E. Kala-azar Krankheit. Tropische Splenomegalie, Eulenburg's Real-Enzyklopädie der Gesamten Heilkunde, vol. 30. New series, vol. 4, pp. 275-277.
1907 —— Trypanosomenkrankheit (Schlafkrankheit) und kala-azar. iii + 52 pp. Jena: G. Fischer.
1907 —— Kala-azar (fieberhafte tropische Splenomegalie) bei einem Schantung-Chinesen. Berliner klinische Wochenschr., vol. 44, pp. 1042-1044.
1909 —— Remarks on piroplasmosis. Philippine Journ. of Sci., April, Section B, Medical Sciences, vol. 4, No. 2, pp. 121-125.
1911 MARZINOWSKY, E. Kultur der Protozoa. Russky Wratch., No. 6. Reprinted in Centralbl. f. Bakt. Culture di Protozoi. Pathologica, November 15, vol. 3, No. 73, pp. 661-662.
1912 —— Maladies voisines de la malaria en Russie, kala-azar, fievre de Malta. Bull. Soc. Path. Exot., December, vol. 5, No. 10, pp. 868-876.
1925 MARZINOWSKY, E. J. Zur Frage der Parasitologie und pathologischen Anatomie der Hundeleishmaniose. Beihefte z. Arch. f. Schiffs.- u. Trop.-Hyg., vol. 20, pp. 234-239.
1911 MARZOCCHI, V. Di un flagellato parassita del tubo digerente del *Ctenocephalus canis* L. Pathologica, June 1, vol. 3, No. 62, pp. 256-257.
1910 MASSAGLIA, ANGELO. Sulla biologia dell' agente spezifico del kala-azar infantile. Società Medico Chirurgia di Modena, Session 1909, December 17, vol. 12. Il Policlinico (sez. pratica), vol. 17, pp. 175. Riforma Medica, vol. 26, pp. 134.
1912 —— Contributo allo studio delle infezioni da *Leishmania infantum*. Nota preventiva. Pathologica, June 1, vol. 4, No. 86, pp. 308-310.
1904 MATHIAS, H. B., and LEISHMAN, W. B. A case of Dum-Dum fever. Journ. Roy. Army Med. Corps, vol. 2, pp. 303-312.
1911 MATHIS, C. Cultures de *Leishmania infantum* and *L. tropica* sur milieux au sang chauffés. Comptes Rendus Soc. Biol., December 8, vol. 71, No. 34, pp. 538-539.
1915 DA MATTA, A. A. Subsidio para o estudo da physionomia clinica classificaçao e synon. das leishmaniosis na America do sul. Brazil Med., September 8, vol. 29, No. 34, pp. 265-268.
1916 —— Tableau synoptique de la classification des leishmanioses. Bull. Soc. Path. Exot., December, vol. 9, No. 10, pp. 761-762.
1918 —— Notas a margem sobre classificaçao das leishmanioses. Amazones Medico, vol. 1, Nos. 3-4, pp. 86-92.
1911 MAURO, CIRO. xxvi. Caso di anemia splenica infantile da corpi di Leishman. La Pediatria, January, series 2, vol. 9, No. 1, pp. 48-51.
1925 MAYER, M. Versuche mit Kala-azar-Erregern. Arch. f. Schiffs- u. Trop.-Hyg., vol. 29, pp. 507-513.
1914 MAYER, M., and WERNER, H. Kultur des Kala-azar-Erregers (*Leishmania donovani*) aus dem peripherischen Blut des Menschen. Deutsch. med. Wochenschr., January 8, vol. 40, No. 2.
1918 —— and REINHARD, P. Zwei Fälle von Kala-azar (Leishmaniose) bei Deutschen (aus Nordafrika bzw. Kleinasien.) Deutsch. med. Wochenshr., February 7, vol. 44, No. 6, pp. 150-152.
1926 MAYER, MARTIN. Susceptibility of the European hamster to kala-azar. Arch. f. Schiffs- u. Trop.-Hyg., August, vol. 30, No. 8, pp. 347-348.
1926 MAZZA, S. Phlebotomi in Tabasco and their rôle in transmission of leishmaniasis. Bol. Inst. de Clin. Quir., vol. 2, pp. 310-317.
1926 —— and CORNEJO ARIAS, J. Primeros casos autoctonos de kala-azar infantil comprobados en el norte de la Republica. Bol. Inst. de clin. quir., vol. 2, pp. 140-144. Also: Rev. Soc. argent. de biol., vol. 2, pp. 96-101.
1913 MAZZITELLI, PIETRO. Interno un caso di anemia infantile da leishmania proveniente da Monte S. Biagio in Provincia di Caserta. Policlinico, sez. medica, February, vol. 20, No. 2, pp. 78-87.
1921 MAZZONI, L. Ricerche ematologische in bambini leishmaniotici dopo l'iniezione di adrenalina. Pediatria, April 15, vol. 29, No. 8, pp. 347-359.
1922 MEGAW, J. W. D. A note on a new disease, "Dermal Leishmaniasis" (Brahmachari). Ind. Med. Gazette, April, vol. 57, No. 4, pp. 128.
1925. MELENEY, H. E. The histopathology of kala-azar in the hamster, monkey and man. Amer. Journ. Path., March, vol. 1, pp. 147-167.

1925 —— Kala-azar in China with special reference to its histopathology in experimentally infected hamsters. Proc. Roy. Soc. Med., 1924-25, vol. 18, No. 7 (Sect., Trop. Dis. and Parasitology), pp. 33-40.

1925 —— Über Wucherung des Lungengefässepithels bei mit Kala-azar infizierten Affen. Arch. f. Schiffs- u. Trop.-Hyg., vol. 29, pp. 685-689.

1925 —— Demonstration of experimental kala-azar in the hamster. Proceedings of a meeting of the Society on February 19, 1925, held at 11, Chandos Street, Cavendish Square. Trans. Roy. Soc. Trop. Med. Hyg., vol. 18, No. 8, p. 394.

1926 DE MELLO, F., and BARRETO, F. Contribution a l'étude de la valeur diagnostique de la réaction de Gaté et Papacostas et de quelques autres réactions alliées. Bull. Soc. Path. Exot., vol. 19, pp. 127-133.

1910 MELLO, U. Toxoplasmiasi o kala-azar? Giornale della reale Società naz. ed Accademia veterinaria italiana, Torino, vol. 59, p. 7.

1927 MESSIK, R. E. Ueber Thrombozytobarine gegen Amœba endolimax und *Leishmania tropica*. Cent. f. Bakt., I. Abt., Orig., vol. 101, pp. 413-417.

1925 MICHAEL, D. F. Note on the incidence of kala-azar on the Pusa Estate. Ind. Journ. Med Res., July, vol. 13, pp. 131-139.

1926 —— A study of the incidence of kala-azar in certain parts of North Behar. February, Indian Medical Research Memoir, pp. 277-280.

1913 MIGONE, L. E. Un caso de kala-azar a Asuncion (Paraguay). Bull. Soc. Path. Exot., February, vol. 6, No. 2, pp. 118-120.

1916 MILLAN, J. F. A note on Leishman bodies, pernicious anæmia, cancer. Med. Pres. Circ., June 14, vol. 101, p. 544.

1914 MILHIT, J. Le kala-azar infantile ou Méditerranéen. Gaz. des. Hôp. Civils et Militaires, May 16, vol. 87, No. 57, pp. 931-938.

1921 MILIO, G. Sulla diffusione della leishmaniosi interna a Messina e dintorni. Pediatria, December 15, vol. 29, No. 24, pp. 1109-1119.

1922 —— Su una siero-reazione precipitante nella diagnosi di leishmaniosi. Pediatria, October 15, vol. 30, No. 20, pp. 957-959.

1922 MILIO, DR. JULIUS. On a serum precipitating reaction in the diagnosis of leishmanial diseases (Translation). Ind. Journ. Med., pp. 265-267.

1909 MILLS, F. Sporadic kala-azar in Behar. Ind. Med .Gazette, December, vol. 44, pp. 450-453.

1922 MILLS, P. S. Note on the formalin test as applied to the diagnosis of kala-azar. Ind. Journ. Med. Res., April, vol. 9, No. 4, pp. 847-849.

1916 MINE, N. Kala-azar. Experimental transmission into laboratory animals. Tokyo Igak. Zassi, September 5, vol. 30, No. 17, pp. 12-148. (China Med. Journ.)

1917 —— The experimental transmission of kala-azar to laboratory animals. Sci-t-kwai Med. Journ., November 10, vol. 30, No. 11, whole number 429, pp. 108-109.

1923 MITRA, A. C. Anti-kala-azar campaign and cost of management of a centre. Ind. Med. Rec., September, pp. 251-254.

1923 —— Malaria and kala-azar and measures to be adopted by the District Boards towards their mitigation. Ind. Med. Record, August, pp. 234-235.

1925 MITRA, N. C. Urea stibamine in kala-azar; a report of sixteen cases. Calcutta Med. Journ., 1925-26, vol. 20, pp. 177-183, 1 chart.

1924 MITRA, R. N., and MAITY, B. B. Speedy recovery from kala-azar after intravenous injections of urea stibamine. Ind. Journ. Med., vol. 5, pp. 144.

1926 MONTENEGRO, J. Cutaneous reaction in leishmaniosis. Arch. Dermat. and Syph., vol. 13, pp. 187-194.

1924 —— The inoculability of leishmania. Amer. Journ. Trop. Med., pp. 331-340.

1908 M'KAIG, ANDREW. Atoxyl in the treatment of kala-azar. Edin. Med. Journ., new series 1, pp. 539-540.

1922 MOLLOW, W. Über einen Fall von Kala-azar, behandelt mit "205 Bayer." Arch. f. Schiffs- u. Trop.-Hyg., vol. 26, No. 9, pp. 273-282.

1925 MOS-CHKOWSKY, SCH. Über die Einwirkung von Germanin ("Bayer 205") auf Leishmanien. (Muhlens.) Arch. f. Schiffs- u. Trop.-Hyg., February, vol. 29, No. 1, pp. 40-44.

1908 MUHLENS, F. Über einige fieberhafte Tropenkrankheiten. Berliner klinische Wochenschr., vol. 45, No. 36, pp. 1631-1633.

1926 MUHLENS, P. Ein mit Antimosan geheilter Fall von indischen Kala-azar. Arch. f. Schiffs- u. Trop.-Hyg., vol. 30, No. 6, pp. 160-166.

1911 MUIR, E. Treatment of kala-azar by the hypodermic injection of a solution of quinine sulphate. Ind. Med. Gazette, February, vol. 46, No. 2, pp. 58-60.

1913 —— The diagnosis and treatment of chronic malaria and kala-azar. Ind. Med. Gazette, July, vol. 48, No. 7, pp. 267-268.

1914 MUIR, E. Further notes on the treatment of kala-azar with antimonium tartaratum. Ind. Med. Gazette, October, vol. 51, No. 10, pp. 368-369.
1915 —— Treatment of kala-azar by tartar emetic intravenously. Ind. Med. Gazette, October, vol. 50, No. 10, pp. 365-368.
1917 —— Some further hints on the treatment of kala-azar with antimony tartrate. Ind. Med. Gazette, September, vol. 52, No. 9, pp. 317-319.
1918 —— Kala-azar, its diagnosis and treatment.
1025 MUKHERJEE, A. K. (Correspondence.) A hint on the transmission of kala-azar. Ind. Med. Gaz., September, pp. 451-452.
1923 MUKHERJE, H. N. Calcium content of blood in kala-azar. (A preliminary note.) Calcutta Med. Journ., September, vol. 18, No. 3, pp. 394-396.
1926 —— Studies in kala azar. Cal. Med. Journ., March, pp. 383-388.
1923 MUKHERJI, BIJOY CHANDRA. Measures adopted in Assam for control of kala-azar and suggestions for modification in Bengal. Ind. Med. Record, August, pp. 223-224.
1925 MUKHERJEE, S. K. Newer preparations of antimony in the treatment of kala-azar (Editorial). Ind. Med. Record, March, vol. 45, No. 3, pp. 79-80.
1926 —— The problem of transmission of kala-azar (Editorial). Ind. Med. Record, March, vol. 46, No. 3, pp. 80-81.
1927 MURISON, T. D. Annual Public Health Report of the Province of Assam for the year 1925. Ind. Med. Gazette, January, pp. 52-54.
1927 —— Treatment Campaign against Kala-azar. Assam Health Bulletin, No. 9, Government of India Central Publication Branch, Calcutta.
1925 MURUGESAN, P. Some clinical observations on kala-azar. Ind. Med. Gazette, December, vol. 50, No. 12, pp. 457-458.
1906 MUSGRAVE, W. E., WHERRY, W. B., and WOOLEY, P. G. Tropical splenomegaly. Bull. Johns Hopkins Hosp., vol. 17, No. 178, pp. 28-32.
1892 MYA, G., and TRAMBUSTI, A. Contributo allo studio dell' anemia splenica infantile. Lo Sperimentale, Mem. orig., pp. 350-388.

1923 NANDI, SHAMBHUNATH. Different factors in the control of malaria and kala-azar in Bengal. Ind. Med. Record, September, pp. 251-254.
1921 NAPIER, L. E. Kala-azar: Note on the diagnosis and treatment. Ind. Med. Gazette, November, vol. 56, No. 11, pp. 401-404.
1922 —— Kala-azar: A preliminary note on the treatment by the intramuscular injection of a special preparation of soda antimony tartrate. Ind. Med. Gazette, January, vol. 57, No. 1, pp. 10-16.
1922 —— A new serum test for kala azar. Ind. Journ. Med. Res., April, vol. 9, No. 4, pp. 830-846, and Ind. Med. Gazette, p. 338.
1922 —— A report on the treatment of ten cases of kala-azar by sodium acetyl para-amino phenyl-stibiate (stibenyl). Proc. Roy. Soc. Med. (Section Trop. Diseases and Parasit.), August, vol. 15, No. 10, pp. 44-45.
1922 —— An analysis of the clinical picture in kala-azar. Ind. Med. Gazette, November and December, vol. 57, Nos. 11-12, pp. 406-412, 446-451.
1923 —— Incidence of kala-azar in Bengal. Ind. Med. Gazette, July, vol. 58, No. 7, pp. 299-301.
1923 —— Musings on the problem of the transmission of kala-azar. Calcutta Med. Journ.
1923 —— Treatment of kala-azar by "stibenyl." A report of ten cases. Lancet, February 10, pp. 280-283.
1923 —— Further practical experience with the aldehyde test. Ind. Med. Gazette, March, vol. 58, No. 3, pp. 104-107.
1923 —— The treatment of kala-azar by meta-chlor-para-acetyl-amino-phenyl stibiate of sodium. 11 cases. Ind. Med. Gazette, December, vol. 58, No. 12, pp. 578-582.
1923 —— The failure of "Bayer 205" in the treatment of kala-azar. Ind. Med. Gazette, September, vol. 58, No. 9, pp. 415-417.
1924 —— The reaction of the blood in kala-azar. Ind. Journ. Med. Res., January, vol. 11, No. 3, pp. 719-732.
1924 —— The problem of cure in kala-azar. Ind. Med. Gazette, October, pp. 492-504.
1925 —— A comparative study of the environment associated with kala-azar prevalence in Calcutta. Ind. Journ. Med. Res., April, vol. 12, No. 4, pp. 755-772.
1925 —— A preliminary note on the successful treatment of kala-azar with "stibamine glucoside." Ind. Med. Gazette, vol. 60, pp. 24-26.
1925 —— A new organic antimony compound for the treatment of kala-azar. Ind. Med. Gazette, vol. 60, pp. 571-572.
1925 —— Stibosan (von Heyden "471"). Ind. Med. Gaz., October, vol. 60, No. 10, pp. 466-467.

1926 —— The pentavalent compounds of antimony in the treatment of kala-azar. Ind. Journ. Med. Res., vol. 14, No. 2, October, pp. 263-279.
1926 —— An epidemiological consideration of the transmission of kala-azar in India. February, Indian Medical Research Memoir, pp. 219-265.
1926 —— Reactions following the administration of the pentavalent compounds of antimony. Ind. Med. Gazette, vol. 61, No. 11, pp. 559.
1927 —— —— Kala-azar.
1927 —— The pentavalent compounds of antimony in the treatment of kala-azar. Vol. 2, No. 603 (Von Heyden), Ind. Journ. Med. Res., vol. 15, No. 1 (in the Press).
1927 —— A new serological test for kala-azar. Ind. Med. Gazette, July, pp. 362-365.
1923 —— and DAS GUPTA, B. M. Cultural examination of the urine in kala-azar. Ind. Med. Gazette, November, vol. 58, No. 11, pp. 530-531.
1927 —— —— and FOSTER, P. The control of kala-azar on tea estates. Ind. Med. Gaz., February, pp. 76-80.
1923 —— and MUIR, E. Kala-azar.
1924 —— —— and MURUGESAN, P. The viability of the flagellate stage of *Leishmania donovani* with reference to the hydrogen-ion concentration of its environment. Ind. Journ. Med. Res., April, vol. 11, No. 4, pp. 1219-1226.
1927 —— —— and SMITH, R. O. A. The development of *Leishmania donovani* in the gut of the sand-fly *Phlebotomus papatasii*. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 713-716.
1926 —— —— —— A study of the bionomics of *Phlebotomus argentipes*, with a special reference to the condition in Calcutta. Ind. Journ. Med. Res. Memoir No. 4, pp. 161-172.
1926 —— —— —— Further observations on the feeding of sandflies, *Phlebotomus argentipes*, on cases of kala-azar in Calcutta. February, Indian Medical Research Memoir, pp. 147-153.
1923 NASSO, IVO. La reazione di Brahmachari nella diagnosi della leishmaniosi infantile. Pediatria, March 1, vol. 31, No. 5, pp. 225-229.
1923 —— —— and MALLARDI, M. Stibioresistenza e stibiointolleranza nella cura della leishmaniosi infantile (observazioni cliniche e ricerche sperimentali). Pediatria, January 15, vol. 31, No. 2, pp. 57-67.
1912 NATTAN-LARRIER, L. La coloration des leishmania dans le coupes. Comptes Rendus Soc. Biol., March 22, vol. 72, No. 11, pp. 436-438.
1918 —— Les cirrhoses hépatiques dues au kala-azar. Bull. Acad. de Méd., May 28, series 3, vol. 79, year 82, pp. 402-403.
1926 NAUCK, E. G. Diagnosis of kala-azar. Tung chi Med. Monatschr., vol. 2, pp. 22-28, 28-30.
1904 NEAVE, SHEFFIELD. *Leishmania donovani* in the Sudan. Brit. Med. Journ., May 28, vol. 1, p. 1252.
1909 NEEB, H. M. Twee gevallen van *Leishmania donovani* de Oost-Indischen Archipel. Geneesk. Tijdschr. Nederl-Indië, vol. 40, afl. 6, pp. 790-807.
1921 —— A serious case of infantile leishmaniosis cured by intravenous injections of tartras kalicostibicus. Trans. 4th Cong. Far East. Assoc. Trop. Med., vol. 2, pp. 274-282.
1922 NEWMAN, R. E. U. Note on three cases of infantile kala-azar. Journ. Roy. Army Med. Corps, May, vol. 38, No. 5, pp. 379-381.
1908 NICOLLE, C. Nouvelles acquisitions sur le kala-azar: cultures, inoculations au chien, étiologie. Comptes Rendus de l'Acad. des Sci., vol. 146, pp. 498-499.
1908 —— Origine canine du kala-azar. Arch. Inst. Pasteur d. Tunis, April, pp. 51-68.
1908 —— Sur trois cas d'infection splénique infantile à corps de Leishman observés en Tunisie. Arch. Inst. Pasteur d. Tunis, February, Part 1, pp. 3-26.
1908 —— Culture des corps de Leishman isolés de la rate dans trois cas d'anémie splénique infantile. Bull. Soc. Path. Exot., February 12, vol. 1, No. 2, pp. 121-126.
1908 —— Reproduction expérimentale du kala-azar chez le chien. Origine canine probable de cette affection. Bull. Soc. Path. Exot., March 11, vol. 1, No. 3, pp. 188-190.
1908 —— Reproduction expérimentale du kala-azar. Origine canine probable de cette infection. Bull. Soc. Sci. Méd. de Tunis, May 11, pp. 112-115.
1908 —— Quelques faits nouveaux relatifs au kala-azar infantile. Bull. Soc. Path. Exot., December 9, vol. 1, No. 10, pp. 602-605.
1909 —— —— Le kala-azar infantile. Ann. de l'Inst. Pasteur, vol. 23, No. 5, pp. 361-401, and No. 6, pp. 441-471.
1909 —— —— Quelques données nouvelles relatives au kala-azar infantile. Bull. Soc. Path. Exot., October 13, vol. 2, No. 8, pp. 457-459 (1910). July 13, vol. 3, No. 7, pp. 431-432.

1910 NICOLLE, C. Etat actuel de la question du kala-azar infantile. Bull. Méd. de l'Algérie, vol. 21, pp. 638-642.
1911 —— Sur les leishmanioses. Revue d'Hyg. et de Police sanitaire, Paris, April, vol. 33, No. 4, pp. 340-357.
1912 —— Origine et rapports du kala-azar et du bouton d'orient. Report presented on September 27, 1912, to the 8th Section of the XV Congress of Hyg. and Demography at Washington. Arch. Inst. Pasteur Tunis, No. 4, pp. 219-224.
1912 —— Statistique des trente premières observations tunisiennes de kala-azar. Arch. Inst. Pasteur Tunis, Part 2, pp. 65-67.
1914 —— Chronique du kala-azar en Tunisie. Bull. Soc. Path. Exot., June, vol. 7, No. 6, pp. 479-481.
1914 —— Aperçu sur le kala-azar. Presse Méd., March 18, No. 22, pp. 213-214.
1916 —— Aperçu sur le kala-azar. Presse Méd., March, vol. 9, No. 3, pp. 126-129; and Arch. Inst. Pasteur, April 1, vol. 9, No. 3, pp. 176-179.
1916 —— Chronique du kala-azar en Tunisie. Bull. Soc. Path. Exot., March, vol. 9, No. 3, pp. 126-129.
1916 —— Chronique du kala-azar en Tunisie. Arch. Inst. Pasteur de Tunis, April 1, vol. 9, No. 3, pp. 176-179.
1917 —— Chronique du kala-azar en Tunisie. Arch Inst. Pasteur Tunis, October, vol. 10, Nos. 1-2, pp. 90-93.
1917 —— Chronique du kala-azar en Tunisie. Bull. Soc. Path. Exot., October, vol. 10, No. 8, pp. 715-719.
1918 —— A propos de la technique de la ponction de la rate. C. R. Soc. Biol., December, No. 10.
1919 —— Chronique du kala-azar en Tunisie pendant l'année 1918. Kala-azar humain. Arch. Inst. Pasteur Tunis, June, vol. 2, No. 1, pp. 41-45.
1921 —— Chronique du kala-azar en Tunisie. Arch. Inst. Past. de l'Afrique du Nord., March, vol. 1, No. 1, pp. 33-39.
1921 —— Addendum au mémoire: Chronique du kala-azar en Tunisie. Arch. Inst. Past. de l'Afrique du Nord, September, vol. 1, No. 3, p. 345.
1925 —— Chronique du kala-azar en Tunis. Arch. Inst. Pasteur de Tunis, January, vol. 14, No. 1, pp. 136-139.
1925 —— and ANDERSON, CH. Conservation du virus de la leishmaniose canine sur les chiens dans les laboratoires. Bull. Soc. Path. Exot., vol. 10, March 14, No. 3, pp. 171-173.
1923 —— —— Recherches expérimentales sur le mode de transmission du kala-azar. Arch. Inst. Pasteur de Tunis, July, vol. 12, No. 2, pp. 168-198.
1923 —— —— Recherches expérimentales sur le mode de transmission du kala-azar Arch. Inst. Pasteur Tunis, July, vol. 14, No. 3, pp. 264-277.
1924 —— —— Recherches expérimentales sur le mode de transmission du kala-azar. Deuxième mémoire. Arch. Inst. Pasteur de Tunis, June, vol. 13, No. 2, pp. 155-164.
1925 —— —— L'immunité dans le kala-azar expérimentale du chien avec quelques données sur l'évolution de la maladie chez cet animal. Immunité naturelle et immunité par première atteinte naturelle. Arch. Inst. Pasteur de Tunis, vol. 14, pp. 278-287.
1926 —— —— Experiments on the mode of transmission of kala-azar. Arch. Inst. Pasteur de Tunis, June, vol. 15, No. 2, pp. 114-117.
1912 —— and BLAIZOT, L. Virulence des cultures de *Leishmania infantum*. Sensibilité du chacal au virus du kala-azar tunisien. Bull. Soc. Path. Exot., November, vol. 5, No. 9, pp. 721-723; Arch. de l'Inst. Past., No. 4, pp. 225-226.
1907 —— and CASSUTO. Sur un cas de kala-azar (splénomégalie tropicale) observé en Tunisie. Bull. Acad. Méd., October 1, p. 263.
1908 —— —— Infection splénique infantile à corps de Leishman-Donovan. Les rapports avec le kala-azar et l'anémie infantile. Presse Médicale, vol. 16, February 8, No. 12, pp. 89-91.
1910 —— and CONOR, A. Application du "606" au traitement du kala-azar (Première note). Bull. Soc. Path. Exot., December 14, vol. 3, No. 10, pp. 717-718.
1912 —— —— Quelques expériences pratiquées avec le virus de leishmaniose naturelle du Chien. Reproduction de la maladie chez le singe. Bull. Soc. Path. Exot., June, vol. 5, No. 6, pp. 351-355.
1914 —— —— Difficulté de conservation du virus de la leishmaniose canine par les passages. Bull. Soc. Path. Exot., June, vol. 7, No. 6, pp. 481-484.
1918 —— and LEBAILLY, C. Multiplicate des rates et éruption peritonéale de tissu splénique chez un chien infecté de kala-azar par inoculation de produits spléniques humains. C. R. Soc. Biol., March 9, vol. 81, No. 5, pp. 231-232.

1912 — — and LEVY. Contribution à la quinzième observation tunisienne de kala-azar; guérison définitive. Arch. Inst. Pasteur de Tunis, Part 2, p. 64.

1911 —— —— Un cas de kala-azar terminé par la guérison. Bull. de la Soc. de Path. Exot., 8 Mars, vol. 4, No. 3, pp. 138-140.

1922 —— CALAMIDA, F., MACOTTA, VILLAN, G., and SPEZZANFUMO. Chronique du kala-azar en Tunisie. LX and LXI. Observations tunisiennes de kala-azar (résumé) LXII tunisien, et traité par le trepol. Observations d'un cas de kala-azar contracté en Sicile et reconnu en Tunisie. Arch. Inst. Past. de l'Afrique du Nord, vol. 2, June, No. 2, pp. 230-239.

1908 —— COMTE, C., and MANCEAUX, L. Recherches sur le kala-azar (nouvelle série d'expériences) :—

1. Virus et cultures. By C. Nicolle.
2. Kala-azar expérimental du chien. By C. Nicolle and C. Comte.
3. Reproduction expérimentale du kala-azar chez le singe (*Macacus sinicus*). By C. Nicolle and L. Manceaux.
4. Animaux réfractaires. By C. Nicolle and L. Manceaux.
5. Origine canine du kala-azar. By C. Nicolle and C. Comte.
6. Présence des corps de Leishman dans le sang périphérique des animaux infectés. By C. Nicolle and C. Comte.
7. Mise au point. Résultats obtenus. By C. Nicolle, Arch. Inst. Pasteur Tunis, July, Part 3, pp. 97-116.

1908 —— CONSEIL, E., COMTE, C., and CASSUTO, E. Recherches sur le kala azar entreprises à l'Institut Pasteur de Tunis.

1. Quatrième observation tunisienne d'anémie splénique infantile à corps de Leishman. By C. Nicolle and E. Conseil.
2. Isolement et culture des corps de Leishman. By C. Nicolle.
3. Reproduction expérimentale du kala-azar chez le chien avec le virus humain. By C. Nicolle.
4. Origine canine du kala-azar. By C. Nicolle and C. Comte.
5. Reproduction expérimentale du kala-azar chez le singe (*Macacus sinicus*). By C. Nicolle.
6. Cinquième observation tunisienne de kala-azar infantile (reconstituée après décès). By E. Cassuto. Étude expérimentale de ce cas et réflexions. By C. Nicolle. Arch. Inst. Pasteur de Tunis, April, Part 2, pp. 51-68.

1908 —— COMTE, C., MANCEAUX, L., and CORTESI, A. Recherches sur le kala-azar infantile entreprises à l'Institut Pasteur de Tunis. Nouvelle série de faits et d'expériences.

1. Sixième observation tunisienne de kala azar infantile. By A. Cortesi.
2. Septième observation tunisienne de kala-azar infantile. By A. Cortesi.
3. Virus et passages. By C. Nicolle.
4. Kala azar expérimental du chien. By C. Nicolle and C. Comte.
5. Kala azar expérimentale du singe. By C. Nicolle and L. Manceaux.
6. La ponction du foie et l'examen du sang périphérique comme moyens de diagnostic du kala-azar infantile ou expérimental pendant la vie. By C. Nicolle and L. Manceaux.
7. Erratum. Arch. Inst. Pasteur de Tunis, October, Part 4, pp. 143-160.

1909 —— CALAMIDA, ORTONA, C., JAEGGY, E., CORTESI, A., LEVY, E. and A., MANCEAUX, L., and COMTE, C. Recherches sur le kala azar infantile de Tunisie entreprises à l'Institut Pasteur de Tunis. Arch. Inst. Pasteur de Tunis, November, Part 4, pp. 174-201.

1911 —— CORTESI, A., and LEVY, E. Application de l'arseno-benzol au traitement du kala-azar de l'enfant. Bull. Soc. Path. Exot., April 12, vol. 4, No. 4, pp. 187-189.

1909 — — GAVIOLO, M., MANCEAUX, L., and COMTE, C. Recherches sur le kala-azar infantile entreprises à l'Institut Pasteur de Tunis. Arch. Inst. Pasteur de Tunis, July, Part 3, pp. 129-138.

1911 —— MORPURGE, L., MARA, E., CORTESI, A., LEVY, E., CONOR, A., and CONSELL, E. Nouveaux faits d'observation ou d'expérience rélatifs au kala-azar. Archives de l'Institut Pasteur de Tunis, pp. 111-125.

1924 NOGUCHI, HIDEYO. Action of certain biological, chemical and physical agents upon cultures of leishmania. Some observations on plant and insect herpetomonads. Proc. Internat. Conference on Health Problems in Trop. America, pp. 455-478, with 4 plates.

1925 — — and LINDENBERG, ADOLPH. The isolation and maintenance of leishmania on the medium employed for the cultivation of organisms of the leptospira group of spirochætes. Amer. Journ. Trop. Med., January, vol. 5, No. 1, pp. 63-67, with 1 plate.

1926 NOGUCHI, H., and TILDEN, EVELYN B. Comparative studies of herpetomonads and leishmania. 1. Cultivation of herpetomonads from insects and plants, Journ. Exper. Med., Sept. 1, vol. 44, No. 3, pp. 307-325. 2. Differentiation of the organisms by serological reactions and fermentation tests, *Ibid.*, pp. 327-337.
1927 NORRIE, F. H. B. A case of dermatitis exfoliata cured by injection of urea stibamine (Brahmachari). Ind. Med. Gaz., March, pp. 142-143.
1908 NOVY, F. G. Successful canine infection with cultures of *Leishmania infantum*, C. Nicolle. Journ. Amer. Med. Assoc., October 24, vol. 51, pp. 1423-24; Proc. Soc. Ser. Experimentale Biol. Med., December 1, vol. 6, pp. 26-27.
1900 —— Sur leishmania infantum. Bull. de la Société de Path. Exot., Paris, July 21, vol. 2, No. 7, pp. 385-387.

1910 OLPP, G. Beiträge zur Medizin in China mit besonderer Berücksichtigung der Tropenpathologie. Arch. für Schiffs- und Tropen-Hygiene, vol. 14, 5 Parts, pp. 117-200. (Kala-azar, p. 214.)
1918 OLSEN, O. Serologische Untersuchungen bei zwei Fällen von Kala-azar. Arch. für Schiffs und Tropen-Hygiene, March, vol. 22, No. 6, pp. 81-80.

1926 PALIT, S. K. The treatment of kala-azar with special reference to splenic enlargement. Cal. Med. Journ., May, pp. 465-468.
1922 PANAYOTATOU, A. Quelques cas de splénonégale et de kala-azar autochtones chez les indigènes en Egypte. Bull. Soc. Path. Exot., vol. 15, November 8, No. 9, pp. 843-853.
1923 PANJA, GANAPATI. Prevention of malaria and kala azar in Bengal. Ind. Med. Record, September, pp. 244-246.
1912 PANTO, V. La leishmaniose spontanea del cane a Catania. Gazetta degli Ospedali e delle Cliniche, March 5, vol. 33, pp. 289-290.
1925 PARADISO, F. Due casi di kala-azar infantile a sintomatologia ed associazioni morbose non frequenti. Pediatria, vol. 33, pp. 1228-1236.
1925 —— Infantile kala-azar with unusual symptoms and complications. Pediatria, vol. 33, pp. 1228-1236.
1926 —— Sulla distribuzione altimetrica del kala-azar infantile in provincia di Catania e sulla età a die piccoli leishmaniotici. Pediatria, Riv., vol. 34, pp. 664-668.
1926 —— The distribution in altitude of infantile kala-azar in Catania and the age of the children affected. Pediatria, June 15, vol. 34, No. 12, pp. 664-668.
1882 PARISSIS, N. P., and TETZIS, J. A. La maladie endémique des enfants à Hydra appelée Tzanaki. In: De l'île d'Hydra (Grèce), au point du vue méd. et particulièrement du Tzanaki, vol. 8, pp. 33-64.
1927 PARROT, L., and DONATIEN, A. Leishmaniose cutanée primitive expérimentale de la souris blanche. Compt. Rend. Soc. de Biol., vol. 96, pp. 448-449.
1925 —— and LESTOQUARD, F. Sur quelques détails de la structure des leishmania. Bull. Soc. Path. Exot., vol. 18, pp. 541-546.
1925 —— —— Details of the structure of leishmania. Arch. Inst. Pasteur d'Algérie, December, vol. 3, No. 4, pp. 327-332.
1919 PASTORE, R. Chemotherapia nella leishmaniosi interna. Osservazione cliniche. Pediatria, February, vol. 27, No. 2, pp. 96-107.
1914 PATANE, C. Sulla trasmissibilità della *Leishmania infantum* ai Topi Albini. Boll. Accad. Scienze Naturali Catania, May, series 2, No. 31, pp. 62-66.
1917 —— Sul primo caso autoctono di leishmaniosi interna in Cirenaica. Pathologica, July 1, vol. 9, No. 207, pp. 181-183.
1926 PATARINO, G. B. Epidemiology and statistics. Pediatria, vol. 34, pp. 1323-1325.
1926 —— Kala-azar at Bari. Pediatria, Dec. 1, vol. 34, No. 23, pp. 1323-1325.
1914 PATER, H. El kala-azar infantil. Rev. Espec. Méd., Madrid, vol. 17, pp. 808-814.
1907 PATTON, W. S. The development of the Leishman-Donovan parasite in *Cimex rotundatus*. Appendix to the annual report upon the work of the Bacteriological Section of the King Institute of Preventive Medicine, Guindy, for the year 1907.
1907 —— Preliminary report on the development of the Leishman Donovan body in the bed-bug. Scientific memoirs by officers of the Medical and Sanitary Departments of the Government of India. New series, No. 27, pp. 1-19.
1907 —— The development of the Leishman-Donovan parasite in *Cimex rotundatus*. Scientific memoirs by officers of the Medical and Sanitary Departments of the Government of India. New series, No. 31, pp. 1-18.
1908 —— *Herpetomonas lygaei*. Arch. für Protistenkunde, vol. 13. Comparison of the parasite with that of kala-azar, pp. 12-14.
1908 —— Inoculation of dogs with the parasite of kala-azar (*Herpetomonas donovani*) with some remarks on the genus *Herpetomonas*. Parasitology, December 1, No. 4, pp. 311-313.

1909 —— The parasite of kala-azar and allied organisms. With discussion: Dr. Sambon, Dr. Low, Sir Havelock Charles, Dr. Wenyon, Sir Patrick Manson and Captain Patton. Trans. Soc. Trop. Med. and Hyg., 1908-1909, vol. 2, pp. 113-141. Journ. Trop. Med. and Hyg., March 15, vol. 12, No. 6, pp. 87-91.

1909 —— The parasite of kala-azar and allied organisms. Lancet, January 30, vol. 1, No. 5, pp. 306-309. Brit. Med. Journ., January 23, vol. 1, p. 216 (Abstract).

1909 - - - A critical review of our present knowledge of the hæmo-flagellates and allied forms. Parasitology, May-June, vol. 2, Nos. 1-2, pp. 91-139. Leishmania, p. 126.

1912 - - - The development of the parasite of Indian kala-azar (*Herpetomonas donovani*, Laveran and Mesnil) in *Cimex rotundatus* Sign. and in *Cimex lectularius* Linn., with some observations on the behaviour of the parasite in *Conorhinus rubrofasciatus* de Geer. Scientific memoirs by officers of the Medical and Sanitary Departments of the Government of India. New series, No. 53, p. 38.

1912 - - The kala-azar problem. Brit. Med. Journ., November 2, No. 2705, pp. 1194-1196.

1913 - - - Further observations on the development of *Herpetomonas donovani* in *Cimex rotundatus* and *Cimex lectularius*. Proceedings of the Third Meeting of the General Malaria Committee held at Madras, November 18, 19 and 20, 1912, pp. 221 232; and Sci. Mem. Inst., No. 52, p. 38.

1913 —— Is kala-azar in Madras of animal origin? Ind. Journ. Med. Res., July, vol. 1, No. 1, pp. 185-195, and Proceedings of the Third Meeting of the General Malaria Commission held at Madras, November 18, 19, 20, 1912, pp. 215-220 (Government Central Branch Press, Simla, 1913).

1914 - - - The behaviour of the parasite of Indian kala-azar in the dog-flea, *Ctenocephalus felis* Bouche, with some remarks on canine kala-azar and its relation to the human disease. Ind. Journ. Med. Res., July, vol. 2, No. 1, pp. 399-403.

1914 - - - The examination of peripheral blood of 84 patients suffering from kala-azar at the General Hospital, Madras, during the period from June 15 to July 15, 1913. Ind. Journ. Med. Res., October, vol. 2, No. 2, pp. 492-504.

1922 - - Some reflections on the kala-azar and oriental sore problems. Ind. Journ. Med. Res., January, vol. 9, No. 3, pp. 496-532.

1926 - - and HINDLE, EDWARD. Notes on kala-azar in Shantung. Proc. Roy. Soc., Oct. 1, ser. B, vol. 100, No. B, 704, pp. 379-384.

1926 —— —— Notes on the species of sandflies (genus *Phlebotomus*) of North China. Proc Roy. Soc., series B, vol. C, No. 704, p. 405.

1927 —— —— The development of Chinese leishmania in *Phlebotomus major* var. *chinensis* and *P. sergenti* var. Proc. Roy. Soc., May 2, vol. 101, ser. B, No. B 710, pp. 369-390.

1921 - - - LA FRENAIS, H. M., and SUNDARA RAO. Studies on flagellates of the genera *Herpetomonas*, *Crithidia* and *Rhynchoidomonas*. Ind. Journ. Med. Res., October, vol. 9, No. 2, pp. 240-251.

1927 —— —— The development of Chinese leishmania in *Phlebotomus major* var. *chinensis* and *P. sergenti* var. Proc. Roy. Soc., ser. B, pp. 369-390.

1914 PAVONI, G. Contributo alla diagnosi biologica della leishmaniosi. Lavori d. Soc. Italiana di Patologia Esotica, pp. 65-68 Mal. e Malat. d. Paesi Caldi, September-December, vol. 5, Nos. 5-6, pp. 364-367.

1915 —— Contributo alla studio della infezione sperimentale del *Mus musculus* con *Leishmania tropica* and *infantum*. Pathologica, March 1, vol. 7, No. 152, pp. 114-116.

1893 PEDIATRIA, LA, Naples. Stato attuale degli studi sull' anemia splenica infettiva dei bambini, vol. 1, pp. 3-6.

1913 PEDROSO, ALEX M. Leishmaniose local do cao. Local leishmaniosis of dog. Annaes Paulistas de Med. e Cirurgia, September, vol. 1, No. 2, pp. 33-39.

1921 —— Biologia da leishmania. Bol. Soc. Med. e Cirrurg. de S. Paulo, Brazil, May, vol. 4, series No. 2; No. 3, pp. 33-34.

1922 PERRY, H. MARRIAN. Some observations on the occurrence of leishmania in the intestinal tissues in Indian kala-azar, on the pathological changes occasioned by their presence, and on their possible significance in this situation. Journ. Roy. Army Med. Corps, November, vol. 39, No. 5, pp. 323-329.

1910 PERSENAIRE, J. B. C. Kala azar in Ned.-Indië. Medische Revue, Haarlem, vol. 10, pp. 161-165.

1905 PETRONE, G. A. Sulle anemie nell' infanzia. Atti di Relazione al v. Congresso Italiano di Pediatria. Il Policlinico (sezione pratica), vol. 12, Part 24, pp. 749-750.

1910 —— Anemia splenica e anemia leucemica infantile. Il Policlinico (sezione pratica), vol. 17, Part 1, pp. 13-14.

1912 PETRONE, G. A. Sopra sette casi d'anemia da leishmania. Riforma Medica, November 30, vol. 28, No. 48, pp. 1322-1328.
1912 —— Un caso di guarigione di anemia leishmania. La Pediatria, November 30, vol. 20, No. 11 (2nd series, vol. 10), pp. 852-854.
1911 PETTIT, AUGUSTE. Transformation lymphoïde du foie au cours des trypanosomiases et de la leishmaniose. Arch. Internat. Pharm. et de Thérapie, Bruxelles and Paris, vol. 21, Parts 3-4, pp. 163-188.
1912 PETROV, N. Caso di splenomegalia tropicale (kala-azar). Società russa di Patalogia. Session of February 25. Ref. in Pathologica, vol. 4, No. 87, p. 361.
1912 PETROW, N. W. Sluchai tropicheskoi splenomegali (kala-azar). Russk. Vrach, No. 26.
1912 —— Ein Fall von Kala-Azar. Virch. Arch. f. Path. Anat. u. Physiol., September 7, vol. 209, No. 3, pp. 453-456.
1910 PFLUGHOFT. Zur Behandlung mit Arsazetin (kala-azar). Münch. med. Woch., p. 1305.
1904 PHILLIPS, LLEWELLYN. Note on the occurrence of the Leishman-Donovan parasite in Arabia and Egypt. Journ. Trop. Med. and Hyg., vol. 7, pp. 236-237. Discussion, p. 262.
1904 —— Note on the occurrence of the Leishman-Donovan parasite in Arabia and Egypt. Brit. Med. Journ., July 23, vol. 2, p. 195 and p. 657.
1905 PIANESE, G. Sull' anemia splenica infantile. 2° Reunione dei Patalogi in Roma, May, Gazzetta internationale di Medicina, vol. 8.
1908 —— Ulteriori ricerche sull' anemia infantum a leishmania. Atti della reale Accademia Medico-Chirurgico di Napoli, May 31, vol. 3. (Session of March 8), No. 2, p. 16.
1909 — Anemia splenica infantile (con dimostrazione di preparati). Gazzetta degli ospedali, Milan, vol. 30, No. 73, pp. 780 781.
1909 —— Le due forme di anemia splenica infantile. Il Policlinico, Session pratica, vol. 16, No. 24, pp. 754 755.
1909 — Caratteri clinici e reperti ematologici e istopatologici onde si differenzia l'anemia infantum a leishmania (Pianese) de l'anemia infantum pseudoleucemica (Jaksch). Atti della reale Accademia Medico Chirurgico di Napoli, No. 1, p. 26.
1802 —— and GIANTURCO. Sull' anemia splenica infantile. Gazzetta degli ospedali, Milan, vol. 13, No. 130, p. 1284.
1911 PICCIONE, M. Contributo clinico allo studio dell' anemia splenica infantile (kala-azar). Medicina nuova, vol. 2, pp. 395-307.
1912 PITTALUGA, G. El kala-azar infantil (esplenomegalia). Parasitaria des los Niños en la Costa de Levante de España. Rivista clinica de Madrid, October 1, p. 7.
1914 —— Kala azar infantile e leishmaniosi canina in Ispagna. Pathologica, March 1, vol. 6, No. 128, pp. 121-123.
1925 — Leishmaniose en Espagne. Publication of the League of Nations. C. H., 308, Geneva.
1926 —— Epidemiological study of leishmaniasis visceralis in Spain. Journ. Trop. Med. Hyg., December 1, vol. 29, No. 23, pp. 387 398.
1912 —— DIESTRO, J. G., and VILA, M. Estudios sobre el "kala-azar infantil" y la "Leishmania infantum" en España. Bol. Inst. Nac. de Hyg. de Alfonso XIII, December, No. 32, pp. 17-45.
1916 PORCELLI TITONE, F. L'azione antiriproduttiva dei raggi ultra-violetti studiata sui protozoi (leishmania). Pediatria, March, vol. 24, No. 3, pp. 147-151.
1908 POROT. Sur les anémies pseudoleucémiques infantiles à propos d'une observation. Bull. Soc. des Sci. de Tunis, May 11, pp. 89-91. Extract in Arch. Inst. Pasteur de Tunis, July, Part 3, pp. 139-141.
1898 POWELL, A. Prevalence of certain intestinal parasites in India, with some remarks on kala-azar. Ind. Med. Gaz., vol. 33, pp. 441 443.
1905 PRAG, ANDERS. Den tropiska splenomegaliens parasit. Hygiea, vol. 67, pp. 1215-1221.
1910 PRASHAD, D. N. Kala-azar in Patna. Ind. Med. Gaz., August, vol. 45, pp. 295-296.
1902 PRICE, J. DODDS. Notes on kala-azar. Ind. Med. Gaz., vol. 37, p. 370.
1917 —— Notes on an anomalous type of kala-azar. Ind. Med. Gaz., December, vol. 52, No. 12, pp. 427-429.
1920 —— Antimony in kala-azar. Brit. Med. Journ., September 18, pp. 453-454.
1920 —— Kala-azar in Europeans in the Nowgong District of Assam. Ind. Med. Gazette, March, vol. 55, No. 3, pp. 87 89.

1923 —— Thirty years' experience of kala-azar in the Nowgong district of Assam. Ind. Med. Gaz., July, vol. 58, No. 7, pp. 296-299.
1924 —— Cases of kala-azar showing little or no improvement with sodium antimony tartrate subsequently cured by urea stibamine. Ind. Med. Gaz., September, vol. 59, No. 9, pp. 464-466.
1914 —— and ROGERS, L. The uniform success of segregation, measures in eradicating kala-azar from Assam tea gardens. Its bearing on the probable mode of infection. Brit. Med. Journ., February 7, pp. 285-289.
1925 —— and STRICKLAND, C. The significance of the splenic index in kala-azar endemic areas. Ind. Journ. Med. Res., 1925 26, vol. 13, pp. 1-6.
1914 PRINGAULT, E. La leishmaniose canine à Marseilles. Bull. Soc. Path. Exot., June, vol. 7, No. 6, pp. 484-488.
1914 —— Existence de la leishmaniose canine à Marseilles. Bull. Soc. Path. Exot., January, vol. 7, No. 1, pp. 41-42.
1910 PUECH, R. O disodo-luargol na therapeutica das manifestaçoes da heredo syphilis e da leishmania. Ann. Paulist. Med. e Cirurg., December, vol. 10, No. 12, pp. 268-276.
1910 PULVIRENTI, G. Sulla cultura della leishmania. Atti dell' Accademia Gioenia di Scienze Naturali in Catania. 5ª serie, vol. 3, mem. 18, 4p., 1 pl.
1911 —— La leishmaniosi del cane a Catania. Pathologica, May 1, vol. 3, No. 60, pp. 205-206.
1913 —— Anatomia patologica, diagnosi, prognosi e cura. Mal. e Malat. d. Paesi Caldi, October-December, vol. 4, Nos. 6, 7, 8, pp. 350-367.
1910 —— and TOMASELLI, A. Sulla trasmissibilità della leishmania di Catania. Pathologica, vol. 2.
1926 PUPO, J. DE A. Tratamento da leishmaniose da mucosa pelo "eparseno" (amino-arseno-phenol. de Pomret). (Estudo preliminar.) Brazil Medico, vol. 1, pp. 201-204. Also: Sciencia Medica, vol. 4, No. 5; in Portuguese, pp. 207-211, in French, pp. 212-216.

1913 QUILICHINI. Un cas de leishmaniose infantil suivi de guérison. Formules leucocytaires dans la leishmaniose. Bull. Soc. Path. Exot., July, vol. 6, No. 7, pp. 495-498.

1909 RACH, E., and ZAREL, MAX. Über den Kulturellen Befund bei dem in Wien beobachteten Fall von Kala-azar. Deutsches Archiv für klinische Medizin, Leipzig, vol. 96, pp. 387-396, mit 1 Tafel. Münchener medizinische Wochenschrift, vol. 56, p. 387.
1909 DE RAADT, O. L. E. Het voorkomen van Kala-azar of Tropische splenomegalie in Nederlandsch Indië. Geneesk. Tijd. v. Ned.-Indië, Batavia, vol. 49, 6th edition, pp. 750-783.
1926 RAMAN, T. K. Kala-azar. Madras Med. Coll. Mag., vol. 5, pp. 185-194.
1927 —— Kala-azar in Madras. Madras Med. Coll. Mag., March, vol. 6, No. 4, pp. 273-294.
1914 RANIERI, G. Leishmaniosi umana e canina a campo Calabro. Lavori d. Soc. Italiana de Patologia Esotica, p. 115.
1921 RAVANT, PAUL. Deux cas de leishmaniose cutanée contractée en Espagne et en France. Premier cas de contagion en France. Arch. de Dermat. et de Syph., series 6, vol. 2, No. 1, pp. 20-37.
1921 RAY, CHARUBRATA. Hæmolytic test in kala-azar. Ind. Med. Gaz., January, vol. 56, No. 1, pp. 9-10.
1924 —— The globulin content of the serum in kala-azar. Ind. Med. Gaz., August, vol. 59, No. 8, pp. 387-391.
1926 —— Further observations on the hæmolytic test for early diagnosis of kala-azar. Ind. Journ. Med., vol. 7, Part 3, September, pp. 103-108.
1927 —— Globulins in kala-azar and syphilis. Cal. Med. Journ., September, vol. 22, No. 3, pp. 115-124.
1927 RAY, CHOWDHURY AMAL K. Kala-azar.
1914 RAYBUD, A. Les leishmanioses. Marseilles Méd., vol. 51, p. 580. (Index Medicus.)
1925 RAYNAUD, L. Leishmaniosis in North Africa. Report of the Health Section of the Secretariat, League of Nations. Monthly Epidemiol., vol. 4, pp. 609-615.
1914 REED, A. C. Kala-azar. A case report from China. Journ. Amer. Med. Assoc., October 31, vol. 63, No. 18, pp. 1572-1573.
1921 REMLINGER, P. Un cas de kala-azar infantile observé au Maroc. Arch. Inst. Peste de l'Afrique du Nord, Sept., vol. 1, No. 3, pp. 240-241.

1922 RENAULT, JULES, MONIER-VINARD, and GENDRON, G. Kala-azar infantile d'origine française. Guérison par l'acétyl-p-amino-phenyl-stibanate de Soude (stibenyl). Bull. et Mém. Soc. Méd. Hôp. de Paris, December 7, Year 38, third series, No. 34, pp. 1624-1632.

1926 REPORTS OF KALA-AZAR COMMISSION, INDIA. Ind. Journ. Med. Res., Report No. 1, 1924-25 (Indian Medical Research Memoirs, No. 4), pp. 19-280.

1925 REPORT OF THE 2ND ALL BENGAL KALA-AZAR CONFERENCE. Opened by H.E. Lord Lytton, the Governor of Bengal, held at the Government House, Calcutta, on November 21, 1925; the Rotunda Hall, Writers' Building, Calcutta, on November 22, 23, and 24, 1925, pp. iv and 161, with 12 figs, published by Secretary, The Central Co-operative Anti-Malarial Society, Ltd.

1925 REZENDE, M. O., DE. Cura da leishmaniose das mucosas. Ann. Paulist. Med. e Cirurg., vol. 16, pp. 135-142.

1914 RHO, F. Le varie forme di leishmaniosi e le loro specie o varietà parassitarie. Ann. di Med. Nav. e Colon, April-May, Ann. 20, vol. 1, Nos. 4-5, pp. 429-436.

1920 RISIQUE, CEBRIAN R., and VARGAS PESADO, A. M. Estudios Medicos de Murcia.

1910 ROBERT, L. Un cas de kala-azar observé à Madagascar. Ann. d'Hyg. et de Méd. colon., No. 4, pp. 750-758.

1911 ROBERTSON, A. WHITE. Haemateikona. The significance of the blood picture in disease. Ind. Med. Gaz., May, vol. 46, No. 5, pp. 161-175. Kala-azar, p. 164.

1925 RODRIGUES, S. L. A plea for immuno-therapy in kala-azar. Cal. Med. Journ., 1925-26, vol. 20, pp. 421-431.

1897 ROGERS, L. The lower Bengal Burdwan epidemic fever reviewed and compared with the present Assam epidemic malarial fever (kala-azar). Ind. Med. Gaz., vol. 32, pp. 401-408.

1897 —— Report of an investigation of the epidemic of malarial fevers in Assam, or kala-azar. Shillong. Printed at the Assam Secretariat Printing Office.

1898 —— The epidemic malarial fever of Assam, or kala azar. A reply to criticisms. Ind. Med. Gaz., vol. 33, pp. 210-213 and 246-253.

1898 —— On the epidemic malarial fever of Assam, or kala-azar. Medio-Chirurg. Trans., vol. 81, pp. 241-257; Brit. Med. Journ., vol. 1, pp. 819-820.

1898 —— The epidemic malarial fever of Assam, or kala-azar, successfully eradicated from tea-garden lines. Brit. Med. Journ., vol. 2, pp. 891-892; Ind. Med. Rec., (1899), vol. 16, pp. 517-519.

1899 —— The epidemic malarial fever of Assam, or kala-azar (Abstract). Brit. Med. Journ., vol. 2, p.1464; Lancet, vol. 1, p. 1633; Proc. Roy. Med. Chirurg. Soc., 3rd series, vol. 11. p. 142.

1899 —— The results of segregation of cases and moving from infected sites in eradicating the Assam epidemic malarial fever, or kala-azar. Medico-Chirurg. Trans., vol. 82, pp. 395-309.

1902 —— Note on the serum reactions and the temperature curve in chronic malaria, including kala-azar. Ind. Med. Gaz., vol. 37, pp. 377-379.

1904 —— Note on the occurrence of Leishman Donovan bodies in "cachexial fevers," including kala-azar. Brit. Med. Journ., vol. 1, pp. 1249-1251.

1904 —— Cachexial fever in India associated with Cunningham-Leishman-Donovan bodies. Brit. Med. Journ., vol. 2, pp. 645-650; Journ. Trop. Med. and Hyg., August 15, p. 259.

1904 —— On the development of flagellated organisms (trypanosomes) from the spleen protozoic parasites of cachexial fevers and kala-azar. Quart. Journ. Micros. Sci., November, vol. 48, new series, No. 191, pp. 367-377.

1904 —— Leishman-Donovan bodies in malarial cachexia and kala-azar. Ind. Med. Gaz., vol. 39, p. 158; and Brit. Med. Journ., vol. 1, p. 1240.

1904 —— Preliminary note on the development of trypanosoma in cultures of the Cunningham-Leishman-Donovan bodies of cachexial fever and kala-azar. Lancet, vol. 2, July 3, pp. 215-216.

1905 —— The conditions affecting the development of flagellated organisms from Leishman bodies and their bearing on the probable mode of infection. Lancet, June 3, vol. 1, pp. 1484-1487.

1905 —— The nature and prophylaxis of the fevers in the Dinajpur district. Ind. Med. Gaz., vol. 40, pp. 90-95. Kala-dukh and kala-azar, pp. 91-92.

1905 —— The diagnostic and prognostic value of the leucopenia in cachexial fever and kala-azar and its treatment by quinine and bone-marrow. Brit. Med. Journ., April, vol. 1, pp. 705-710.

1906 —— Further work on the development of the herpetomonas of kala-azar and cachexial fever from Leishman-Donovan bodies. Proc. Roy. Soc. B., vol. 77, No. B. 517, pp. 284-293.

1907 —— The Milroy lectures on kala-azar, its differentiation and its epidemiology (abstracts). Brit. Med. Journ., vol. 1, pp. 427-433, 490-494, 557-562. Lancet, vol. 1, pp. 486-491, 568-572, 643-648.

1907 —— Kala-azar. Journ. Roy. Inst. Pub. Health, vol. 15, pp. 227-229.
1908 —— A peculiar intralobular cirrhosis of the liver produced by the protozoal parasite of kala-azar. Ann. Trop. Med. and Parasit., vol. 2, No. 3, pp. 147-152.
1910 - - - Fevers in the tropics. 2nd edition, Oxford University Press, London.
1914 —— The bearing of Assam tea-garden experience on the problem of ætiology of kala-azar. Ind. Journ. Med. Res., vol. 5, pp. 15-20.
1915 —— A preliminary note on the treatment of kala-azar by tartar emetic intravenously and inunctions of metallic antimony. Ind. Med. Gaz., October, vol. 50, No. 10, pp. 364-365.
1915 —— Further work on the treatment of kala-azar, with special reference to leucocyte-increasing methods, spleen tabloids and alkalies. Ind. Med. Gaz., May, vol. 50, No. 5, pp. 163-170.
1915 —— Tartar emetic in kala-azar. (Correspondence.) Brit. Med. Journ., July 31, p. 197.
1916 —— Further cases of kala-azar in Europeans successfully treated with intravenous injection of tartar emetic. Lancet, No. 4, pp. 782-785.
1917 —— Chronic splenomegaly in Lower Bengal, with special reference to prevalence and clinical differentiation of kala-azar. Ind. Med. Gaz., January, vol. 52, No. 1, pp. 7-15.
1917 —— Further experience in the tartar emetic treatment of kala-azar, including its use in young children. Ind. Med. Gaz., July-August, vol. 52, Nos. 7-8, pp. 241-244, 265-269.
1918 —— Sodium antimonyl tartrate in kala-azar. Ind. Med. Gaz., May, vol. 53, No. 5, pp. 161-164.
1919 —— Fevers in the tropics. Third edition, Oxford Medical Publications.
1919 —— Colloid antimony sulphide intravenously in kala-azar, with a note on antimony oxide orally. Lancet, March 29, pp. 505-506.
1915 —— and SHORTEN, A. J. The alkalinity of the blood in kala-azar and cholera and the technique of its estimation. Ind. Journ. Med. Res., April, vol. 2, No. 4, pp. 867-881.
1916 —— and HUME, N. H. Treatment of kala-azar (Indian form) by tartar emetic intravenously and by inunctions of metallic antimony. Brit. Med. Journ., February 26, pp. 301-303.
1914 ROIG-RAVENTOS. Kala azar infantil. Rev. de Cien. Méd. de Barcel., vol. 40, pp. 481-496.
1915 —— Tratamiento del kala azar infantil. Rev. de Cien. Méd. de Barcel., vol. 41, pp. 145-147. (Index Medicus.)
1899 ROSS, RONALD. Report on the nature of kala-azar. Calcutta: Office of the Superintendent of Government Printing.
1899 —— Infectiousness of malarial fever and kala-azar. Ind. Med. Gaz. (extract), vol. 34, pp. 233-241.
1903 —— Note on the bodies recently described by Leishman and Donovan. Brit. Med. Journ., November 14, vol. 2, pp. 1261-1262. (See also p. 1359.)
1903 —— Further notes on Leishman bodies (illustrated). Brit. Med. Journ., vol. 2, p. 1401.
1903 —— A new parasite of man. Thomson Yates and Johnston Laboratories Reports, December, vol. 5 (new series), Part 2, pp. 79-82.
1904 —— The Leishman-Donovan body found at Omdurman. Brit. Med. Journ., vol. 1, p. 1049.
1904 —— *Leishmania donovani* found in kala-azar. Brit. Med. Journ., vol. 1, p. 160.
1904 —— Trypanosomes and the Leishman-Donovan bodies. Brit. Med. Journ., July 9, vol. 2, p. 98.
1913 ROUX, F. Arsenic in the treatment of kala-azar (abstract). Ind. Med. Gaz., April, vol. 48, No. 4, pp. 132-133.
1889 ROUX, M. Traité pratique des maladies des pays chauds. Vol. 3, p. 263; and 2nd edition, vol. 1, p. 529.
1911 - - - Sur un cas de leishmaniose observé à Alger. Bulletin de l'Académie de Médecine, Paris, 6 Juin, 3e série, vol. 65, No. 23, p. 644.
1904 ROW, M. C. N. Guaiaquin: A remedy suggested for piroplasmosis, kala-azar, and other allied fevers. Ind. Med. Gaz., vol. 34, pp. 455-456.
1909 ROW, R. Evolution of the diagnostic methods in kala-azar, with a special reference to the technique for intensive culture from the patient's finger blood. Trans. Grant. Coll. Med. Soc., p. 8.
1913 —— Some experimental facts *re* kala-azar. Journ. Trop. Med. and Hyg., January 1, vol. 16, No. 1, pp. 1-2.
1912 —— *Leishmania donovani* and *Leishmania tropica*. Brit. Med. Journ., March 30, No. 2674, pp. 717-718.

1912 ROW, R. Kala-azar. Brit. Med. Journ. (Correspondence), March 30, No. 2674, p. 758.
1912 —— A simple haemoglobinized saline culture medium for the growth of leishmania and allied protozoa. Brit. Med. Journ., May 18, No. 2681, pp. 1119-1120.
1912 —— Some experimental facts *re* kala-azar (Indian). Brit. Med. Journ., November 2, p. 1196; Journ. Trop. Med. and Hyg., November 1, vol. 15, No. 21, pp. 327-328.
1914 —— Experimental leishmaniasis in the monkey and the mouse induced by the parasites in culture. Ind. Journ. Med. Res., April, vol. 1, No. 4, pp. 617-621.
1922 —— On reversion of the flagellate form of *Leishmania donovani* and *Leishmania tropica* to the resistant non-flagellate torpedo and O body, in culture tubes, and its bearing on the attempts at the search for the transmitter. Ind Journ. Med. Res., October, vol. 10, No. 2, pp. 476-481.
1924 —— Some pathological observations in experimental leishmaniasis in mice, with special reference to generalized infections set up by *Leishmania tropica*. Ind. Journ. Med. Res., vol. 12, No. 2, pp. 435-438.
1925 —— Canine leishmaniasis in Bombay. Ind. Med. Gaz., July, vol. 60, No. 7, pp. 317-318, with 3 text figs.
1923 ROY, R. D. Kala-azar in Malda and some practical difficulties in its treatment Ind. Med. Rec., September, pp. 243-244.
1923 ROY, D. N. Personal observations on a few cases of kala-azar. Ind. Med. Gaz., January, vol. 58, No. 1, pp. 23-24.
1926 ROZIER, L. Notes sur la leishmaniose canine. Bull. Soc. Path. Exot., vol. 19, pp. 179-182.
1904 RUATA, G. R. Kala-azar o splenomegalie tropicale. Clinica Medica Italiana, June, p. 418. Abstract in Arch. Gén. de Méd., 2e semestre, p. 2426.
1904 —— Kala-azar, or tropical splenomegaly. Journ. Trop. Med. and Hyg. (extract), vol. 7, pp. 350-352.
1925 RUGE, PROF. DR. VON REINHOLD, MUHLENS, PROF. DR. PETER, and VERTH, PROF. DR. MAXZUR. Krankheiten und Hygiene der Warmen Lender.
1914 RUTELLI, G. Sulla ricerca dei parassiti di Leishman nel sangue periferico (della leishmania interna). Lavori d. Soc. Italiana di Patologia Esotica, pp. 60-61.
1914 —— Infezione sperimentale della *Leishmania umana* nei topolini. Lavori d. Soc. Italiana di Patalogia Esotica, pp. 64-65.

1915 SAITO. The clinical observation of kala-azar in a Chinese child. Sei-i-Kwai Med. Journ., January 10, vol. 34, No. 1 (whole No. 395). The original in No. 22, vol. 22 of the Journ. Tokio Med. Assoc.
1913 SALVATORE, DOMENICO. Un caso di kala-azar a Derna. Mal. e Malat. d. Paesi Caldi, March, vol. 4, No. 2, pp. 73-76.
1914 —— Colture di *Leishmania hominis* iniettate nel peritoneo dei cani. Mal. e Malat. d. Paesi Caldi, February, vol. 5, No. 1, pp. 29-32.
1905 SAMBON, L. W. Kala-azar. Climate, vol. 6, pp. 245-250.
1922 SAMUT, ROBERT. Colloidal antimony in *Leishmania infantum*. Lancet, May 20, p. 995.
1910 SANGIORGI, G. Sulla possibilità della trasmissione dei protozoi, parassiti del sangue per mezzo del *Cimex lectularius*. Giorn. della reale Accad. di Med. di Torino, 4 series, vol. 16, pp. 228-232.
1911 —— Ancora sulla presenza di forme di leishmania nel *Pulex serraticeps*. Pathologica, March 1, vol. 3, No. 56, pp. 89-90.
1911 —— A proposito dell' articolo del Dr. Franchini, "La *Leishmania donovani* può vivere e svilupparasi nell' intestino dell' Anopheles." Pathologica, December 1, vol. 3, No. 74, pp. 691-692.
1911 —— Leishmaniosi spontanea dei cani a Torino. Pathologica, December 15, vol. 3, No. 75, pp. 699-700.
1911 —— Lettere all' Editore. Pathologica.
1911 —— Sulla presenza di forme di *Leishmania infantum* (Nicolle) nella pulce (*Pulex seraticeps*) dei cani randaggi di Catania. Pathologica, January 15, vol. 3, No. 53, pp. 23-24. Il Policlinico (Sezione pratica), vol. 18, Part 7, pp. 209-210.
1923 SANGIVA, RAO. B. Kala-azar. Madras Med. Journ., January-March, p. 56.
1926 SANYAL, C. C. Observations on a few kala-azar cases. Cal. Med. Journ., August, pp. 55-60.
1927 —— Rectal administration of antimony for the treatment of kala-azar. Cal. Med. Journ., February, pp. 405-408.
1927 —— Two cases of confirmed malaria followed up to development cured by injection of antimony. Cal. Med. Journ., February, pp. 428-429.

1927 —— Several cases of antimony intolerance. Cal. Med. Journ., February, vol. 21, No. 8, pp. 424-426.
1915 SARKER, S. L. The action of quinine and arsenical preparations in kala-azar. Ind. Med. Gaz., March, vol. 50, No. 3, pp. 92-94.
1923 —— Kala-azar-infected villages in Malda District. Ind. Med. Rec., August, pp. 208-214.
1923 —— and DE, A. N. Some observations on the aldehyde test. Calcutta Med. Journ., December, vol. 18, No. 6, pp. 530-543.
1911 SAVILLE, L. E. Correspondence on kala-azar infantum in China. Further notes. China Med. Journ., July, vol. 25, No. 4, pp. 273-274.
1898 SCHEUBE, B. Ponos. Eulenburg's Realenzyklopädie der gesammten Heilkunde, 3 aufl. XIX, pp. 318-319.
1903 —— Kala-azar. Ebenda, 3 aufl., vol. 27 (neue folge 1), pp. 405-406.
1903 —— The diseases of warm countries. A handbook for medical men. Translated by P. Falcke, edited by J. Cantlie, 8 vols. London: John Bale, Sons and Danielsson, Ltd. (Kala-azar, pp. 149-150; Ponos, pp. 307-308; Oriental Sore, pp. 534-540.)
1910 —— Die Krankheiten der warmen Länder im Handbuch für Ärzte. Vierte umgearbeitete und erweiterte Auflage. Pp. viii + 1072, with 5 maps, 1 plate and 142 figures in text. Jena: Verlag von Gustav Fischer. (Kala-azar, pp. 211-225; Ponos, pp. 586-588; Die endemische Beulenkrankheit, pp. 962-973.)
1927 SCHLEGELMILCH, WALTER. Über Kala-azar. Aus dem für Schiffs- und Tropenkrankheiten zu Hamburg.
1925 SCHILLING, V. Zur Frage der Einschlusskörper (Chlamydozoa von Prowazek). II. "Schollenleukozyten" in entzündlichen. Beihefte z. Arch. f. Schiffs- u. Trop.-Hyg., vol. 29, pp. 322-326.
1920 SCHMIDT, HANS. Die Pharmakosynthese organischer Antimonverbindungen. Pharm. Ztg., 1920, No. 89, p. 872.
1922 —— Das Antimon in der neueren Medizin. Beihefte z. Arch. f. Schiffs- u. Trop.-Hyg.
1909 SCHNEIDER, G. E. Leishmanioses. Gazette des hôpitaux civils et militaires, Paris, vol. 132, pp. 1543-1547.
1910 —— La leishmaniose infantile. La Clinique infantile. February, année 8.
1924 SCHULZ, MARTIN. Die Kernteilung von *Leptomonas fasciculata* nebsteinem Vergleich mit der Kernteilung bei *Leishmania donovani*. (Nuclear division of *L. fasciculata* and its comparison with the same process in *L. donovani*. Arch. f. Protistenk., Nov. 5, vol. 49, No. 2, pp. 216-236, with 1 plate.
1910 SCORDO, FRANCESCO. Prime ricerche sul ricambio materiale in un caso di kala-azar. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia. Roma: Tipografia Labicana, vol. 2, pp. 22-38.
1910 —— Contributo alla conoscenza della patologia dei reni e delle capsule surrenali nel kala azar. Studio intorno ad alcune malattie tropicali della Calabria e della Sicilia. Roma: Tipografia Labicana, Part 2, pp. 51-54.
1911 —— La vitalità della *Leishmania donovani* in cultura ed in contatto coi batteri del tubo digerente delle pulci e delle cimici. Pathologica, September 15, Anno, 3, No. 69, pp. 457-458.
1911 —— Prime ricerche sul ricambio materiale in un caso di kala-azar. Policlinico, Oct. 22, vol. 18, No. 43.
1912 —— I leucociti della cavia e del coniglio in contatto delle forme flagellate della *Leishmania donovani* "in vitro" e nel corpo degli animali. Mal. e Malat. d. Paesi Caldi, September-October, vol. 3, Nos. 9-10, pp. 240-249.
1912 —— Die Vitalität der *Leishmania donovani* in Berührung mit dem Bakterien des Verdauungstraktus der Flöhe und Wanzen. Zentralbl. für Bakt., Ie Abt. Orig., April 6, Part 1, vol. lxiii, pp. 62-64.
1913 —— Über einige Infektionsversuche der "Anopheles" mit dem Milzsaft von Leishmaniosiskranken. Zentralbl. f. Bakt., 1 Abt. Orig., July 29, vol. 70, Nos. 1-2, pp. 36-41.
1913 —— Über die Frage nach der Übertragbarkeit des Kala-azar durch einige blutsaugende Insekten. Zentralbl. f. Bakt., 1 Abt. Orig., August 23, vol. 70, Nos. 5-6, pp. 307-319.
1913 —— Alterazione morfologiche dei corpi del Leishman nel kala-azar. Mal. e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 313-317.
1913 —— A proposito di alcuni tentativi d'infezione delle "Anopheles" con succo splenico di malattie di leishmaniosi interna. Mal. e Malat. d. Paesi Caldi, March, vol. 4, No. 2, pp. 84-89.
1913 —— Die Leukocyten des Meerschweinchens und des Kaninchens in Kontakt mit den Flagellatenformen der *Leishmania donovani* in vitro und im Körper der Tiere. Zentralbl. f. Bakt., 1 Abt. Orig., May 3, vol. 69, Nos. 1-2, pp. 85-80.

1913 SCORDO, F. Sulla questione della trasmissibilità del kala-azar per mezzo di alcuni insetti ematofagi. Mal. e Malat. d. Paesi Caldi, January, vol. 4, No. 1, pp. 20-32.

1914 —— Sulla pretesa identità della *Leishmania hominis* e della *Leishmania canis.* Mal. e Malat. d. Paesi Caldi, July-August, vol. 5, No. 4, pp. 265-271.

1914 —— Intorno alla natura di certi corpi granulari recentemente descritti nella leishmaniosi. Mal. e. Malat. d. Paesi Caldi, July-August, vol. 5, No. 4, pp. 272-276.

1909 SCOTT, P. W. The parasite of kala-azar and allied organisms. Journ. Trop. Med. and Hyg., vol. 12, pp. 87-91.

1926 SEN GUPTA, TARAPADA. Unusual symptoms following the administration of urea stibamine. Ind. Med. Gazette, vol. 61, No. 8, August, pp. 394-395.

1910 SERGENT, ED. and ET. Kala-azar. Existence de la leishmaniose chez les chiens d'Alger. Bull. Soc. Path. Exot., vol. 3, No. 8, pp. 510-511.

1921 —— Formes leishmaniennes et leptomonadiennes chez les punaises de Chauves-souris. C. R. Soc. Biol., July 23, vol. 85, No. 27, pp. 413-415.

1912 —— L'HERITIER, A., and LEMAIRE, G. Transmission de leishmania de chien à chien par piqûres de *Pulex serraticeps*. Bull. Soc. Path. Exot., vol. 5, No. 8, pp. 505-507.

1912 —— LOMBARD and QUILICHINI. La leishmaniose à Alger. Infection simultanée d'un enfant, d'un chien et d'un chat dans la même habitation. Bull. Soc. Path. Exot., Séance of February 14, vol. 5, No. 2, pp. 93-98.

1916 —— and DE MOUZON. Quatrième observation algérienne de kala-azar. Bull. Soc. Path. Exot., November, vol. 9, No. 9, pp. 694-696.

1926 SERGENT, ET., CATANEI, A., GUEIDON, E., BOUGUET, A., and DES ISLES, H. M. Le clou de Mila. Arch. Inst. Pasteur d'Algerie, vol. 3, pp. 1-8.

1926 SERGENT, E., GUEIDON, E., and PAGES, A. Cases in Algeria. Statistics. Arch. Inst. Pasteur d'Algerie, vol. 4, pp. 26-29.

1922 SEYFARTH, C. Eine einfache Methode zur diagnostischen Entnahme von Knochenmark beim Lebenden. Arch. f. Schiffs- u. Trop.-Hyg., vol. 26, December, No. 11, pp. 337-341.

1923 —— Die Sternumtrepanation, eine einfache Methode zur diagnostischen Entnahme von Knochenmark bei Lebenden. Deut. med. Woch., February 9, vol. 49, No. 6, pp. 180-181.

1920 SHAHA, B. B. Treatment of kala-azar by intramuscular and oral medication. Calcutta Med. Journ., October, p. 14.

1926 SHANKS, G., and KHAN, G. P. The diagnosis of typhoid-like fevers, with special reference to typhoid and kala-azar Ind. Med. Gazette, vol. 61, No. 7, pp. 324.

1923 SHORTT, H. E. Record of kala-azar research work carried out at the King Edward VII Memorial Pasteur Institute, Shillong, during 1922. Ind. Journ. Med. Res., April, vol. 10, No. 4, pp. 1150-1168.

1923 —— *Herpetomonas ctenocephali* Fantham. Some observations on its life history and reactions to different environments. Ind. Journ. Med. Res., vol. 10, No. 3, p. 733

1923 —— The infectivity of insect flagellates to vertebrates, with a special reference to *H. ctenocephali*. Ind. Journ. Med. Res., vol. 10, No. 4, p. 908.

1923 —— The recovery of *Herpetomonas donovani* from urine of a kala-azar patient. Ind. Journ. Med. Res., July, vol. 11, No. 1, pp. 319-320.

1923 —— The pathology of acute experimental kala-azar in monkeys. Ind. Journ. Med. Res., July, vol. 11, No. 1, pp. 186-195.

1924 —— The relative value of diagnostic methods and evidence of cure in kala-azar. Ind. Med.Gaz., November, pp. 551-552.

1924 —— The present position as regards the problem of transmission of kala-azar. Proc. Assam Branch British Medical Assoc. Annual Meeting, Jorhaut, February 16 and 17, pp. 8-18.

1925 —— and BRAHMACHARI, U. N. Chemotherapy of antimonial compound in kala-azar infection. Part III, Further observations on dermal leishmanoid. Ind. Journ. Med. Res., January, No. 13, vol. 12, pp. 463-466.

1926 —— BARRAUD, P. J., and CRAIGHEAD, A. C. Note on a massive infection of the pharynx of *Phlebotomus argentipes* with *Herpetomonas donovani*. Ind. Journ. Med. Res., January, vol. 13, No. 3, pp. 441-443.

1926 —— —— —— *Conorhinus rubrofasciatus* de Geer and Indian kala-azar. Ind. Journ. Med. Res., July, vol. 14, No. 1, pp. 230-242.

1926 —— —— —— The life-history and morphology of *Herpetomonas donovani* in the sand-fly, *Phlebotomus argentipes*. Ind. Journ. Med. Res., April, vol. 13, No. 4, pp. 947-959.

1926 —— —— —— Note on a massive infection of the buccal cavity of *Phlebotomus argentipes* with *Herpetomonas donovani*. Ind. Journ. Med. Res., October, vol. 14, No. 2, pp. 329-330.
1926 —— —— —— The occurrence in nature of *Phlebotomus argentipes* infected with a flagellate morphologically identical with *Herpetomonas donovani*. Ind. Journ. Med. Res., October, vol. 14, No. 2, pp. 521-522.
1926 —— —— —— Note on a massive infection of the pharynx of *Phlebotomus argentipes* with *Herpetomonas donovani*. February, Indian Medical Research Memoir, pp. 157-159.
1926 —— —— —— The finding in nature of *Phlebotomus argentipes* infected with a flagellate identical with *Herpetomonas donovani*. Ind. Journ. Med. Res., vol. 14, No. 2, p. 505.
1927 —— —— —— Note on the infectivity of the forms of *Leishmania donovani* found in *Phlebotomus argentipes*. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 577-579.
1927 —— —— —— Transmission experiments in Indian kala-azar with *Phlebotomus argentipes*. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 589-600.
1927 —— CRAIGHEAD, A. C., KHAZAN CHAND, and SWAMINATH, C. S. The "resistant non-flagellate torpedo and O bodies" of Row seen in old cultures of *Leishmania donovani* in their relationship to the production of infections. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 567-576.
1923 —— and SEN, R. T. Clinical kala-azar work performed at the special kala-azar hospital, Shillong, during 1922. Ind. Med. Gaz., July, vol. 58, No. 7, pp. 289-293.
1924 —— Final report on the use of urea stibamine in kala-azar. Ind. Journ. Med. Res., vol. 12, No. 2, pp. 335-338.
1923 —— —— Urea stibamine in the treatment of kala-azar. Ind. Journ. Med Res., October, vol. 11, No. 2, pp. 653-659.
1923 —— —— and SWAMINATH, C. S. A second report on the recovery of *Herpetomonas donovani* from urine of kala-azar cases. Ind. Journ. Med. Res., October, vol. 11, No. 2, pp. 667-668.
1924 —— and SWAMINATH, C. S. Note on the infection of a mouse by means of bed bugs, *Cimex hemiptera* Fabr., fed on the peripheral blood of a case of kala-azar. Ind. Journ. Med. Res., January, vol. 11, No. 3, pp. 965-966.
1924 —— —— The behaviour of cultures of *Herpetomonas donovani* in bed-bugs, *Cimex lectularius* and *Cimex hemiptera*. Ind. Journ. Med. Res., vol. 12, No. 2, pp. 391-396.
1925 —— —— —— Experiments to decide whether the bed-bug *Cimex hemiptera* Fabr., can transmit Indian kala-azar. Ind. Journ. Med. Res., vol. 13, pp. 143-147.
1925 —— —— Systemic infection of a monkey (*Macacus rhesus*) by intradermal inoculation of spleen puncture material from a case of Indian kala azar. Ind. Journ. Med. Res., p. 149.
1927 —— —— —— The mode of formation and morphology of the O bodies of Row in old cultures of *Leishmania donovani*. Ind. Journ. Med. Res., vol. 14, No. 3, January, pp. 581-587.
1910 SHVETS, YA. Kala azar: black disease: its distribution and relation to Banti's disease. Vrach. gazeta, vol. 17, pp. 703, 731.
1921 SIA, RICHARD H. P. Ray's "haemolytic" test in kala-azar. China Med. Journ., September, vol. 35, No. 5, pp. 397-399.
1924 —— A simple method of estimating quantitative differences in the globulin precipitation test in kala-azar. China Med. Journ., January, vol. 38, No. 1, pp. 35-42.
1921 —— and HSIEN WU. Serum globulin in kala-azar. China Med. Journ., November, vol. 35, No. 6, pp. 527-532.
1913 DA SILVA, P. Notes sur le kala-azar. Arquivos d. Inst. Bact. Cam. Pest., vol. 4, No. 2, pp. 147-172.
1915 —— Experiences sur la transmission de la leishmaniose infantile par les puces (*Pulex irritans*). Arquivos d. Inst. Bact. Cam. Pest., vol. 4, No. 3, pp. 261-267.
1913 SIGNER, M. Sulla distribuzione della leishmaniosi in Italia. Mal. e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 320-323.
1926 SINDONI, M., and GUCCIONE, F. Leishmaniasis in beginning tuberculosis. Pediatria, vol. 34, pp. 1082-1085.
1908 SLUKA, ERICH. Un cas de kala-azar. Semaine médicale, December 2, p. 588.
1909 —— and ZAREL, MAX. Ein Fall von Kala-azar aus Taschkent in Wien. Deut. Arch. f. klin. Med., vol. 96, pp. 356-386, 1 plate.
1909 —— —— —— Ein Fall von Kala-azar. Münch. med. Woch., May, 25th year, vol. 56, pp. 1072-1075.

1913 SMALLMAN, A. B. Note on some cellular bodies found in a case of Mediterranean leishmaniosis. Journ. Roy. Army Med. Corps, December, vol. 21, No. 6, pp. 636-640.
1906 SMITH, JOHN. A case of piroplasmosis. Splenectomy followed in eight months by death. Ind. Med. Gaz., vol. 41, pp. 15-16.
1924 SMYLY, H. J. Experiments on the administration of tartar emetic by various routes. Proc. Soc. Exper. Biol. and Med., vol. 22, pp. 201-203.
1926 —— Chemotherapy of experimental leishmaniasis in hamsters. Trans. of the Roy. Soc. of Trop. Med. and Hyg., March and May, vol. 20, Nos. 1 and 2, pp. 104-110.
1927 —— The administration of tartar emetic by various routes. Annals of Trop. Med. and Parasit., vol. 21, No. 2, July 22.
1924 —— and YOUNG, C. W. The experimental transmission of leishmaniasis to animals. Proc. Soc. Exper. Biol. and Med., 1923-1924, March, vol. 21, No. 6, pp. 354-356.
1926 SOCIÉTÉ DES NATIONS. Organisation d'Hygiène. Étude épidémiologique sur la "Leishmaniose Viscerale" en Espagne. (G. Pittaluga.) (League of Nations. Epidemiological Study of Kala-azar in Spain), pp. 28, with 3 text figs, October 8, Geneva, C. H. 308. In Spanish in Rev. Med. de Barcelona, January, year 3, vol. 5, No. 25, pp. 42-62, with 1 map, and in Italian in Ann. d'Igene, 1926, April, vol. 36, No. 4, pp. 259-284, with 1 map.
1913 SOCIETÀ ITALIANA FRA I CULTORI DELLE MALAT. ESOTICHE. Riunione Privata tenuta a Messina il June 15, 1913, intorno alla leishmaniosi umana in Italia. Atti, relazioni, communicazioni scientifiche contavole per cura del Prof. Dr. G. Spagnolio, Dr. M. Signer, Segretari della Riunione, p. 180. (1) Scordo, F., L'etiologia della leishmaniosi interna nel basino Medit., pp. 17-40. (2) Di Cristina, Sulle sindrome, diagnosi, prognosi e cura dell' anemia da leishmania, pp. 50-62. (3) Gabbi, U., Pellegrino and Montoto, G., Inchiestori intorno al kala-azar nelle Provincie della Sicilia orientale e della Calabria Inf., pp. 63-77. (4) Dionosi, A., Contributo alla anatomia patologica dell'anemia da leishmania, pp. 78-81. (5) Cannata, S., Il sangue nell'anemia de lishmania, pp. 82-92. (6) Idem, Sul reperto del parassita di Leishman nel sangue periferico, pp. 89-92. (7) Caronia, G., L'anafilassi nessa leishmaniosi infantile, pp. 92-93. (8) Idem, Sul potere complem. del siero di sangue nella leishmaniosi infantile, pp. 98-102. (9) Spagnolio, G., Sulla ganglio-puncture nella diagnosi di leishmaniosi, pp. 102-104. (10) Scordo, F., Alterazioni mortol. dei corpi del Leishman nel kala-azar, pp. 105-108. (11) Ranieri, G., Il primo esempio di leishmaniosi interna a campo Calabro, pp. 108-109. (12) Licciardi, S., Sulla leishmaniosi interna in Catania, pp. 110-115. (13) Timpano, P., Un caso di leishmani interna finito con la guarigione, pp. 115-116. (14) Spagnolio, G., Nuovi casi clinico a nicoteia marino ed a nizza Sicilia, p. 117. (15) Vadala, P., Il primo esempio di kala-azar a furnari dimostrato colla splenopuncture, pp. 119-121. (16) Sprandio, G., La cura climatica nella leishmaniosi interna, pp. 121-123. (17) Abate, A., Contributo alla casistica e allo studio della leishmaniosi infantile, pp. 123-126. (18) Idem, La resistanza dei globuli Rossi nella leishmaniosi infant, pp. 126-127. (19) La Cava, Un caso di leishmaniosi interna in una giovinetta di 14 anni, pp. 127-130. (20) Pugliatti, G., Sulla ricorrenza primaverile della leishmaniosi interna, pp. 130-133. (21) Signer, M., Sulla distribuzione della leishmaniosi in Italia, pp. 133-135. (22) Gabbi, U., Sulla identità clinica ed etiologica della leishmaniosi umana e canina, pp. 136-147. (23) Spagnolio, G., Leishmaniosi umana e canina. Studio d'ambiante, pp. 147-152.
1884 SOMMA, L. Sull' anemia splenica infantile. Archivio di patalogia infantile.
1890 —— Dell' anemia splenica infantile; storia, clinica e terapia. Relazione letta al I. Congresso pediatrico Italiano in Roma nell' ottobre dell' anno 1890, Archivio Italiano di Pediatria, vol. 6. Also appears in Allgemeinen Wiener medinizischen Zeitung, vol. 36, pp. 345, 357.
1925 SOULIE, H. La kala azar dans le bassin de la méditerranée. Médecine, 1925-26, vol. 7, pp. 183-186.
1910 SPAGNOLIO, G. Intorno a due nuovi casi di kala-azar in Sicilia. Gazzetta Medica Italiano, vol. 61, p. 201.
1910 —— Intorno a due nuovi casi di kala-azar in Sicilia. Studi intorno ad alcune malattie tropicali in Sicilia e Calabria. Tipografia Labicana, Part 1, p. 44.
1911 —— Nuovi casi di kala-azar nel Comune di Messina. I focolai endemici di Camaro e Casalotto. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestriale redatto da Umberto Gabbi. Tipografia: F. Centenari, Part 1, pp. 31-35. Mal. e Malat. d. Paesi Caldi, vol. 2, No. 2, p. 54.

1911 ——— Intorno alla guarigione spontanea del kala-azar. Studi intorno all malattie tropicali dell' Italia meridionale e insulare e delle Colonie. Archivio trimestrale redatto da Umberto Gabbi, Part 2. Messina: Tipogr., S. Guerriera.
1911 ——— Intorno alla guarigione spontanea del kala-azar. Mal. e Malat. d. Paesi Caldi, August, vol. 2, No. 8, pp. 230 232.
1912 ——— Sulla vitalità della *Leishmania donovani* in simbiosi con i germi delle malattie infettive intestinali e con lo streptococco piogene. Mal. e Malat. d. Paesi Caldi, May-June, vol. 3, Nos. 5-6, pp. 151-152.
1912 ——— Leishmaniosi interna (kala-azar) a Messina nuovi esempi clinici e sguardo riassuntivo. Mal. e Malat. d. Paesi Caldi, November, vol. 3, No. 2, pp. 307-310.
1912 ——— Su alcune modificazioni rilevate nell' equilibrio leucocitario del sangue splenico del kala-azar. Mal. e Malat. d. Paesi Caldi, March, vol. 3, No. 3, pp. 74-75.
1913 ——— Nota clinica su alcuni recenti casi di leishmaniosi interna (kala azar). Mal. a Malat. d. Paesi Caldi, March, vol. 4, No. 2, pp. 80-82; and Riforma Med., May 17, vol. 29, No. 20, pp. 530-538.
1913 ——— Sulla ganglio-puntura nella diagnosi di leishmaniosi. Mal. e Malat. d. Paesi Caldi, August-September, vol. 4, No. 5, pp. 306-308.
1913 ——— Leishmaniosi interna (kala-azar) a Messina. Nuovi esempi clinici e Sguardo riassuntivo. Riforma Med., February 22, vol. 29, No. 8, pp. 190-201.
1913 ——— Sulla coltivabilità della leishmania, cosi detta infantum, nel sangue splenico-citrato o Mezzo di Rogers. Mal. e Malat. d. Paesi Caldi, January, vol. 4, No. 1, pp. 2-7.
1913 ——— Leishmaniosi canina e umana e loro rapporti. Mal. e Malat. d. Paesi Caldi, April-May, vol. 4, No. 3, pp. 203-204.
1914 ——— Le complicanze delle vie respiratorie nella leishmaniosi interna. Lavori d. Soc. Italiana di Patologia Esotica, pp. 83 86.
1914 ——— Leishmaniosi umana e canina. Studio d'ambiente. Ref. Med., February 14, vol. 30, No. 7, pp. 179-182.
1915 ——— Vizio cardiaco e leishmaniosi interna. Mal. e Malat. d. Paesi Caldi, July-August, vol. 6, No. 4, pp. 191-192.
1915 ——— Leishmaniosi canina ed umana e loro presunta dipendenza genetica. Mal. e Malat. d. Paesi Caldi, May June, vol. 6, No. 3, pp. 150-157.
1915 ——— Die Leishmaniosi bei Menschen und Hunden. Studium des Krankheitsgebietes. Zentralbl. f. Bakt. Orig., January 15, vol. 75, No. 4, pp. 294-298.
1916 ——— La cura della leishmaniosi interna con i preparati di antimonio. Mal. e Malat. d. Paesi Caldi, July August, vol. 7, No. 4, pp. 242-245.
1920 ——— La cura del kala-azar infantile con i preparati di antimonio. Giorn. di Clin. Med., Bologna, May, vol. 1, No. 5, pp. 182-186.
1913 ——— and GIUGNI, F. Riunione privata tenuta a Messina il 15 Guigno, 1913, intorno alla leishmaniosi umana in Italia.
1914 ——— Stato presente del problema della trasmissione della leishmaniosi interna nei paesi del bacino Mediterraneo (Rivista Sintetico Critica). Mal. e Malat. d. Paesi Caldi, May-June, vol. 5, No. 3, pp. 204-211; July August, No. 4, pp. 297-305.
1913 SPOLVERINI, L. M. Contributo allo studio della leishmaniosi infantum. Pediatria, September 30, vol. 21, No. 9, pp. 650-668.
1910 SPRAWSON, C. A. Kala-azar in Mesopotamia and its incubation period. Brit Med. Journ., November 22, pp. 667-669.
1905 STATHAM, J. C. B. Preliminary note on the cultivation of the Leishman body. Journ. Roy. Army Med. Corps, vol. 4, pp 13 15.
1905 ——— A case of kala-azar. Journ. Roy. Army Med. Corps, September, vol. 5, pp. 248 262, 366-384.
1907 ——— Kala-azar or Dum-Dum fever. Transvaal Med. Journ., 1906-7, vol. 2, pp. 31-33.
1913 ——— and BUTLER, G. G. Note on certain bodies found by liver puncture in a case of fever associated with splenic enlargement. Journ. Roy. Army Med. Corps, vol. 21, No. 6, pp. 620-635.
1881 STEPHANOS. Le ponos de Spetza et d'Hydra. Gaz. Hebd. Méd. et de Chir., 2nd series, vol. 18, No. 47, p. 750; No. 51, p. 813.
1897 STEPHEN, A. Dr. Roger's report on kala-azar. (Abstract.) Ind. Med. Gaz., vol. 32, pp. 408-413.
1904 STEPHENS, J. W. W. Specimens of Leishman-Donovan bodies, or *Helcosoma tropicum*. Liverpool Medico-Chirurgical Journal, vol. 24, pp. 360-371.
1920 STIGTER, P. L. Kala-azar. Een bijdrage tot de casuistiek. Geneesk. Tijdschr. v. Nederl. Indië, vol. 60, No. 1, pp. 44-48.
1924 STRUTHERS, E. B. Kala-azar. Criterion of cure. China Med. Journ., March, vol. 38, No. 3, pp. 207-212.

1926 STRUTHERS, E. B. The advantages of the organic preparations of antimony in the treatment of kala-azar. A preliminary note. China Med. Journ., September, pp. 849-850.
1927 —— Treatment of kala-azar. China Med. Journ., January, pp. 21-28.
1927 —— "Neostam" stibamine glucoside in the treatment of kala-azar. China Med. Journ., January, vol. 12, No. 1, pp. 21-28.
1927 —— The treatment of kala-azar by stibosan (Heyden "471") and antimosan (Heyden "661"). China Med. Journ., September, vol. 41, No. 9, pp. 755-761.
1924 —— and CH'I CHANG CH'UN. The globulin precip. and formol-gel tests in diagnosis of kala-azar. China Med. Journ., March, vol. 38, No. 3, pp. 203-206.
1927 SUAREZ, F. G. The first case of adult kala-azar in Madrid (Arch. de Medicine Cirurgia y Especialidades, Feb. 5), also referred to in the Journ. of Trop. Med. and Hyg., March 15.
1923 SUR, S. N. Epidemiology of kala-azar in Bengal. Ind. Med. Rec., August, pp. 218-222.
1915 SUTHERLAND, W. D., and MITRA, G. C. The Wassermann reaction in malaria, kala-azar and leprosy. Ind. Journ. Med. Res., April, vol. 2, No. 4, pp. 984-989.
1909 SWABEY, L. W. A fatal case of kala-azar. Journ. Roy. Army Med. Corps, September, vol. 13, No. 3, pp. 294-297.
1904 SWAN, J. G. Case of continued fever with Leishman-Donovan bodies. Brit. Med. Journ., vol. 1, No. 2269, p. 1487.
1921 SYMONS, T. H. Lieut.-Col., I.M.S. Madras Annual Report and Statistics of the Government General Hospital, Madras, for the year 1921.

1923 TANABE, N. On the conditions necessary for the development of *Leishmania donovani* in vitro. Saikingaku Zasshi, Journ. of Bact., June, No. 333. Summarized in Jap. Med. World, 1924, February 15, vol. 4, No. 2, p. 46.
1910 TASHIM, IBRAHIM. Sur l'existence en Tripolitaine du kala azar et de la fièvre méditerranéenne. Bull. Soc. Path. Exot., October 12, vol. 3, No. 8, pp. 511-512.
1910 —— Sur l'existence en Tripolitaine du kala-azar et de la fièvre méditerranéenne. Arch. Inst. Pasteur de Tunis, Part 4, pp. 158-159.
1911 THOMSON, Capt. D. S. B., and MARSHALL, Lt. W. E. Kala-azar commission to investigate the prevalence and cause of the disease in the Eastern Sudan. (1) General Report (by Capt. Thomson). (2) Pathological Report (by Lt. Marshall). Fourth Report of the Wellcome Tropical Research Laboratories at the Gordon Memorial College, Khartoum. Vol. A.—Medical, pp. 143-172.
1921 THOMSON, J. GORDON, and SINTON, J. A. *Leishmania donovani* in cultures recovered from spleen puncture during life and from the bone-marrow of the cadaver. Proc. Roy. Soc. Med., vol. 14 (Sec. Trop. Dis.), June, No. 8, pp. 21-23.
1898 THORNHILL, H. A criticism of Dr. Rogers' report on kala-azar. Ind. Med. Gaz., vol. 33, pp. 50 and 86.
1910 TIMPANO, P. Un caso di anemia splenica infantile da parassiti di Leishman. Riforma Medica, May 2, vol. 26, No. 81, pp. 490-491.
1925 —— Un caso di kala-azar in una donna di 23 anni. Policlinico, Sez. Prat., April 13, vol. 32, No. 15, pp. 526-527.
1926 —— Il 1° caso di leishmaniosi esterna curata col radio. Policlin., sez. pratica, vol. 33, pp. 260-262.
1910 TOMASELLI, A. Studio delle leishmanie nel succo della milza dei bambini affetti da kala-azar. Policlinico, sezione medica, June, vol. 17, Part 6, pp. 240-245.
1910 —— Alcune ricerche sul sangue degli ammalati di kala-azar. Rivista Critica di Clinica Medica, vol. 11, No. 22, pp. 337-340.
1910 —— Morfologia delle leishmanie nel succo splenico di bambini affetti da leishmaniosi. Atti dell' Accad. Giornia di Scienze Naturali in Catania. Series 5, vol. 3, mem. 19, p. 3.
1911 —— Sull' anatomia pathologica del kala-azar sperimentale. Folia clinica chimica et microscopia, April, vol. 3, Part. 8.
1913 —— Le complicazioni della leishmaniosi infantile. Mal. e Malat. d. Paesi Caldi, April-May, vol. 4, No. 3, pp. 180-181.
1925 TORRADEMÉ, JOSÉ. Consideraciones clinicas a proposito de diagnostico y tratamiento del kala-azar infantil en la comarca de Tortosa. La Medicine Ibera, No. 415.
1924 TORRES, C. B. MAGARINOS, and LEAO, A. E. DE AREA. Leishmaniose e blastomycose. (Observacao de um caso de associacao das duas molestias.) Sciencia Medica, May 31, vol. 2, No. 5, pp. 256-259, with 8 figs.
1915 TORRES, OCTAVIO. O valor do tartaro emetico na leishmaniose. Tres Observações. Brazil Medico, April 1, vol. 29, No. 13, pp. 97-101.
1917 —— Distribucão geographica da leishmaniose na Bahia. Ann. Paulist. Med. e Cirurg., February, vol. 8, Year 5, No. 2, pp. 39-43.

1917 —— Observação de um caso de leishmaniose destruidora. Brazil Medico, May 5, vol. 31, No. 18, pp. 151-155.
1920 TOURNIER, ÉLIE. Note sur un cas de kala-azar infantile observé au Gabon. Bull. Soc. Path. Exot., March 10, vol. 13, No. 3, pp. 175-176.
1915 TRANTAS, A. I. Ophthalmological observations. II. Ophthalmoscopic changes in generalised leishmaniasis. Arch. de Méd., December 1 20, vol. 10, Nos. 34, 36, pp. 276-279.
1906 TREUTLEIN. Über Protozoenblutkrankheiten bei Mensch und Tier in Indien und Deutsch-Ostafrika. Münch. med. Woch., May, vol. 53, pp. 855-856.
1923 TRIPPUTI, V. Nota clinica sulla leishmaniosi infantile. Pediatria, November 15, vol. 31, No. 22, pp. 1236-1238.
1926 TURKHUD, D. A., KRISHNAN, K. V., and SEETHARAMA IYER, P. V. An investigation of kala-azar in certain endemic areas in Southern India. Ind. Journ. Med. Res., January, vol. 13, No. 3, pp. 703-747.
1910 TYZZER, ERNEST EDWARD, and WALKER, E. L. A comparative study of *Leishmania infantum* of infantile kala azar and *Leptomonas* (*Herpetomonas*) *ctenocephali* parasitic in the gut of a dog flea. Journ. Med. Res., July, vol. 40, No. 2, pp. 129 176.

1913 UHLENHUTH, P., MELZER, and HÜGEL. Die chemotherapeutische Wirkung von organischen Antimonpräparaten bei Spirochäten- und Trypanosomkrankheiten. Deutsche med. Woch., pp. 303 and 2455.
1924 —— PHILALETHES KUHN and HANS SCHMIDT. Über ein neues trypanozides Antimonkomplexsalz (Heyden "661"). Deutsche med. Woch., No. 38.
1925 —— —— —— Chemotherapeutische Antimonstudien. Arch. f. Schiffs- u. Tropen-Hyg., November, No. 11.

1914 VAGLIO, R. Contributo alla ricerca del Parassita di Leishman nel sangue periferico di bambini affetti da leishmaniosi. Pediatria, September, vol. 22, No. 9, pp. 682-686.
1922 —— Trasmissione della leishmaniosi interna. Pediatria, August 1, vol. 30, No. 15, pp. 728-734.
1927 VARMA, R. L. Berberine sulphate in oriental sore. Ind. Med. Gaz., vol. 62, pp. 84-85.
1927 VASILE, B. Contributo alla cono-cenza dell' infezione associata kala-azar-malaria. Pediatria, April 1, vol. 35, No. 7, pp. 376-380.
1909 VENTURA, CLAUDIA. Considerazioni etiologiche intorno ad un caso di anemia splenica infantile. Gazzetta degli Ospedali e delle Cliniche, vol. 30, No. 30, pp. 313-315.
1908 VERDIER, F. Les leishmanioses. Thèse doctorat. Ollier-Henry and Co., p. 89.
1914 VERRIENTI, P., and RUSSI, P. Contributo allo studio clinico del kala-azar. Riforma Med., September, vol. 30, No. 36, pp. 996-999.
1914 VILA, M. Un caso de kala azar en un adulto. Rev. Clin. Madrid, vol. 12, pp. 435 441.
1915 —— Caracteres de la leishmaniosis canina en Tortosa. Rev. Clin. Madrid, vol. 13, p. 172-174.
1925 VILLAIN, C. Note sur le kala azar du Honan (Chine centrale). Bull. Soc. Path. Exot. (Abstract), Oct. 14, vol. 18, pp. 669-676.
1910 VISENTINI, ARRIGO. Über die Morphologie und den Entwicklungskreis der bei Kranken Kalabriens und Siziliens beobachteten Leishmania. Archiv für Schiffs- u. Trop.-Hyg., vol. 14, Part 4, pp. 1-15.
1910 —— Contributo alla conoscenza del kala-azar in Italia. Studi intorno ad alcune malattie tropicali in Sicilia e Calabria. Tipografia Labicana, Part 1, La Riforma Medica, vol. 26, No. 19, pp. 507-509.
1910 —— Sull' anatomia patologica dell' anemia infettiva da leishmania (kala-azar) osservata in Calabria e Sicilia. Pathologica, December 1, vol. 2, No. 50, pp. 569-574.
1910 —— Sulla morfologia e sul ciclo di sviluppo della leishmania osservata nei malati di Calabria e di Sicilia. First communication. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia. Roma: Tipografia, Labicana. Part 2, pp. 9-20.
1910 —— Sulla distribuzione geografica del "kala-azar" in Italia. Il "kala-azar" nelle puglie e nelle isole eolie. Roma: Tipografia, F. Centenari, Part 3, pp. 50-54.
1910 —— Sulla distribuzione geografica del kala-azar in Italia. Mal. e Malat. d. Paesi Caldi, anno 1, p. 157.

1910 VISENTINI, A. Sull' anatomia patologica del kala-azar osservato in Calabria e Sicilia. Studi intorno ad alcune malattie tropicali della Calabria e della Sicilia. Roma: Tipografia, Labicana, Part 2, pp. 41-50.

1911 —— Una carta geografica della distribuzione della leishmaniosi in Italia. Kala-azar e bottone d'oriente. Con tavola. Studi intorno alle malattie tropicali dell' Italia meridionale e insulare e delle colonie. Archivio trimestrale redatto da Umberto Gabbi. Roma: Tipografia, F. Centenari. Part 1, pp. 28-30; Mal. e Malat. d. Paesi Caldi, anno 2, Part 2.

1912 —— Transmission of leishmaniasis by means of cultures and the mechanism of the natural immunity in rats and guinea-pigs. Quart. Journ. Micros. Sci., December, vol. 58, No. 2 (New Series, No. 230), pp. 373-384.

1912 —— On the morphology of the leishmania of Italian kala-azar. Third communication: Cytological researches on leishmania in cultures. Quart. Journ. Micros. Sci., December, vol. 58, No. 2 (New Series, No. 230), pp. 353-371

1912 —— Mécanisme de l'immunité naturelle du rat et du cobaye à l'égard des cultures de *Leishmania infantum*. Bull. Soc. Path. Exot., June, vol. 5, No. 6, pp. 358-360.

1913 —— La mie ricerche di trasmissione delle leishmaniosi. Lettere all' Editore. Pathologica, December 1, vol. 5, No. 122, p. 734.

1913 —— Ricierche morfologiche, culturali e biologiche sulla leishmania della leishmaniosi spontanea del cane. Rendiconti d. R. Accademia dei Lincei, December 7, vol. 22, series 5, No. 11, pp. 582-587.

1915 —— Il kala-azar (Rivista storico-critica) L'Attualita Med., February, vol. 4, No. 2, pp. 81-107.

1911 VOLPINO, G. Infezione sperimentale da *Leishmania infantum* nella cornea del coniglio. Pathologica, February 1, vol. 3, No. 54, pp. 45-46. Policlinico vol. 18, p. 208

1911 —— Experimentelle Infektion mit *Leishmania infantum* in der Hornhaut des Kaninchens. Central. für Bakt., Orig. 1 Abteilung, September 2, vol. 60, Parts 1-2, pp. 91-92.

1923 WAGENER, EDNA HANNIBAL. A skin reaction to extracts of *Leishmania tropica* and *Leishmania infantum*. Univ. California Public Zool., Dec. 31, vol. 20, No. 22, pp. 477-488, with 1 plate.

1926 —— and KOCH, DOROTHY ANN. The biological relationship of leishmania and certain herpetomonads. Univ. California Public Zool., March 16, vol. 28, No. 20, pp. 365-388.

1924 WAHED, A. K. M. ABDUL. A few practical points in kala-azar. Ind. Med. Rec., December, vol. 44, No. 12, pp. 358-362.

1916 WARD, G. R Kala-azar in soldiers returning from Malta. Lancet, July 1, pp. 16-17.

1892 WALSH, J. H. TULL. Case of kala-azar in an insane under care of Surgeon Captain J. H. Tull Walsh. Notes by W. A. Williams, I.S.A. Ind. Med. Gaz., July, vol. 27, pp. 207-208.

1921 WATERSON, J. A contribution to the knowledge of the bionomics of sandflies. Ann. Trop. Med. and Parasit., vol. 16, No. 1, p. 69.

1911 WELD, Major A. E. A clinical note on two cases of kala-azar treated with salvarsan. Journ. Roy. Army Med. Corps, September, vol. 17, No. 3, pp. 275-277.

1915 —— A short note on the work done in the Military Families Hospital, Malta, during the period from January, 1909, to August, 1914. Journ. Roy. Army Med. Corps, June, vol. 24, No. 6, pp. 579-584.

1911 WENYON, C. M. Reports of the Advisory Committee for the Tropical Diseases Research Fund for the year 1911

1911 —— Leishmania and mosquitoes. Lancet (Correspondence), November 11, vol. 2, pp. 1362-1363.

1911 }
1912 } —— Kala-azar Bulletin. Trop. Dis. Bureau, London.

1912 —— Some recent advances in our knowledge of leishmaniasis. Journ. Lond. School Trop. Med., vol. 1, Part 2, pp. 93-98.

1912 —— Note on the occurrence of herpetomonas in the phlebotomus of Aleppo. Journ. Lond. School Trop. Med., Part 2, p. 98.

1913 —— Experiments on the behaviour of leishmania and allied flagellates in bugs and fleas, with some remarks on previous work. Journ. Lond. School Trop. Med., vol. 2, Part I, pp. 13-26.

1914 —— Kala-azar in Malta, with some remarks on the various leishmaniases. Trans. Soc. Trop. Med. and Hyg., January, vol. 7, No. 3, pp. 97-111.

1914 —— The culture of leishmania from the finger-blood of a case of Indian kala-azar, with some remarks on the nature of certain granular bodies recently described from this disease. Journ. Trop. Med. and Hyg., February 16, vol. 17, No. 4, pp. 49-51.
1915 —— Flagellate forms of *Leishmania donovani* in the tissues of an experimentally infected dog. Journ. Trop. Med. and Hyg., October 1, vol. 18, No. 19, pp. 218-219.
1915 —— Leishmania problems: Observations on a recent contribution to the subject. Journ. Trop. Med. and Hyg., November 1, vol. 18, No. 21, pp. 241-247.
1922 —— Kala-azar and the bed-bug. Lancet, February 25, pp. 400-401
1922 —— Leishmaniasis. A review of recent literature. I. Trop. Dis. Bull., January, vol. 19, No. 1, pp. 1-18.
1922 —— Leishmaniasis. A review of recent literature. II. Trop. Dis. Bull., April, vol. 19, No. 3, pp. 182-193.
1926 —— Protozoology.
1910 WHITE, CHARLES. Notes on a case of kala-azar. Journ. Roy. Army Med. Corps, March, vol. 14, No. 3, pp. 313-315.
1914 WHIPHAM, T. R. Case of kala-azar. Proc. Roy. Soc. Med., vol. 7 (Sec. Study of Dis. Child), pp. 63-64.
1909 WILKINSON. Statistics of kala-azar in the Assam district. (Sanitary Report Eastern Bengal and Assam.) Ind. Med. Gaz., 1910, vol. 45, p. 468.
1892 WILLIAMS, W. A. (Notes on.) Case of kala-azar in an insane. Under care of Surgeon-Captain J. H. Tull Walsh. Ind. Med. Gazette, Calcutta, July, vol. 27, pp. 207-208.
1909 WILLIAMSON, G. A. Is ponos kala-azar? (With discussion.) Brit. Med. Journ., September 18, vol. 2, pp. 781-782, and 1910, February 26, vol. 1, pp. 545-546; Journ. Trop. Med. and Hyg. (Abstract), August 16, vol. 12, No. 6, pp. 242-243; Lancet, September 11, vol. 2, p. 799.
1911 —— Is ponos kala-azar? Brit. Med. Journ., January 28, vol. 1, No. 2613, p. 229.
1925 WOLLSTEIN, M. A case of kala-azar in an infant. New York State Journ. Med., March 13, vol. 25, pp. 413-417.
1906 WOOLEY, PAUL G. Tropical febrile splenomegaly. Philippine Journ. of Science. Section B.- Medical Sciences, Manila, June, vol. 1, No. 5, pp. 533-545.
1906 —— *Bacterium pneumoniæ simile* (novo var.). An organism isolated from a case of tropical febrile splenomegaly. Zentralbl. für Bakteriologie, 1 Abt. Orig., vol. 42, pp. 589-590.
1906 WOLFF, H. Über Pathogenese und Therapie der Anæmia splenica infantum. Berliner klinische Wochenschrift, vol. 43, No. 40, pp. 1565-1569.
1924 WOLTRING, F. J. L., and HULK, J. F. Een geval van kala-azar. Ned. Tijdschr. v. Geneesk. August 30, No. 9, pp. 1096-1112.
1906 WURTZ, R. Kala-azar. Nouveau traité de médecine et de Thérapeutique, Brouardel and Gilbert. Maladies exotiques, vol. 6, pp. 228-230. Libraire J. B. Baillière et fils.
1920 WYLIE, J. H. Kala-azar in North China. China Med. Journ., November, vol. 34, No. 6, pp. 593-601.
1925 WYLLIE, W. C. A case of kala-azar. Proc. Roy. Soc. Med. (Sect. Study of Disease in Children), November, vol. 19, pp. 5-6.

1883 XANTHOS, M. G. Sur le tsanaki, maladie de l'enfance sévissant dans l'île d'Hydra. Congrès des Méd. Grec., 1882, vol. 1, p. 79.

1915 YAKIMOFF, W. L. Contribution à l'étude des leishmanioses de l'homme et du chien dans le Turkestan russe. Bull. Soc. Path. Exot., July, vol. 8, No. 7, pp. 474-503.
1915 —— De la période d'incubation chez les animaux infectés par les leishmania. Bull. Soc. Path. Exot., July, vol. 8, No. 7, pp. 430-431.
1923 —— Studie sul kala-azar. Pediatria, August 1, vol. 31, No. 15, pp. 811-816.
1911 —— and KOHL-YAKIMOFF. Leishmaniose canine à Tunis. Bull. Soc. Path. Exot., July 12, vol. 4, No. 7, pp. 452-453.
1912 —— —— Infection des souris blanches par les cultures de *Leishmania infantum* Ch. Nicolle (Travail du laboratorie biologique du George Speyerhaus à Frankfort sur le Mein). Bull. Soc. Path. Exot., séance of April 10, vol. 4, No. 4, pp. 218-220.
1912 —— —— L'infection des animaux de laboratoire par la *Leishmania infantum* Ch. Nicolle. (Deuxième note preliminaire.) Bull. Soc. Path. Exot., June, vol. 5, No. 6, pp. 355-357.

1913 YAKIMOFF, W. L., KOHL-YAKIMOFF, and SCHOKHAR, N. I. Leishmaniose canine à Taschkent. Bull. Soc. Path. Exot., June, vol. 6, No. 6, pp. 432-433.
1914 —— and SCHOKHAR, N. I. Recherches sur les maladies tropicales humaines et animales au Turkestan. 1.—Répartition de la leishmaniose canine au Turkestan. Bull. Soc. Path. Exot., March, vol. 7, No. 3, p. 185.
1923 YORKE, WARRINGTON. The treatment of kala-azar by "Bayer 205." Brit. Med. Journ., March 3, p. 370.
1923 YOUNG, CHARLES W. Kala-azar in China. China Med. Journ., October, vol. 37, No. 10, pp. 797-822.
1925 —— and HERTIG, M. The development of flagellates in Chinese sand-flies (*Phlebotomus*) fed on hamsters infected with *Leishmania donovani*. Proc. Soc. Exper. Biol. and Med., 1925 26, vol. 23, pp. 611-615.
1926 —— —— A search for field and house rodents naturally infected with kala-azar. Proc. Soc. Exper. Biol. and Med., 1925-26, February, vol. 23, pp. 305-308.
1926 —— —— Attempts to transmit kala-azar by means of rodent lice. Proc. Soc. Exper. Biol. and Med., February, vol. 23, pp. 398-402.
1926 —— Attempts to transmit kala-azar by means of bed-bugs (*Cimex* sp.). Proc. Soc. Exper. Biol. and Med., vol. 23, pp. 402-405.
1926 —— and PAO-YUNG LIU. Susceptibility of field, house and laboratory rodents of infection with *Leishmania donovani*. Proc. Soc. Exper. Biol. and Med., February, vol. 23, pp. 302-305.
1922 —— and VAN SANT, H. M. The diagnosis of kala-azar by blood-culture. Proc. Soc. Exper. Biol. and Med., March 15, vol. 19, No. 6, pp. 299-302.
1923 —— —— *Leishmania donovani* in the peripheral blood. Proc. Soc. Exper. Biol. and Med., No. 4, pp. 219-222.
1923 —— —— *Leishmania donovani* in the peripheral blood. Journ. Exper. Med., September 1, vol. 38, No. 3, pp. 233-256.
1924 —— SMYLY, H. J., and BROWN, C. Experimental kala-azar in a hamster (*Cricetulus griseus*). Proc. Soc. Exper. Biol. and Med., March, vol. 21, No. 6, pp. 357-359.
1926 —— —— —— Experimental kala-azar in a hamster, *Cricetulus griseus* M. Edw. Amer. Journ. Hyg., March, vol. 6, pp. 254-275.
1913 YOUNG, T. C. MCCOMBIE. An account of an investigation of the prevalence of endemic kala-azar in the plains of Assam. Proc. 3rd Meeting Genl. Mal. Com. (Madras), November 18-20, 1912, pp. 257-265, Government Central Branch Press, Simla, 1913.
1914 —— Report on the progress of the kala-azar investigation during the session 1912-13. Ind. Journ. Med. Res. supplement, vol. 5, pp. 21-44.
1919 —— A supplement to Sanitary Report of the Province of Assam for 1919. Assam Secretariat Printing Office.
1921 —— Annual Public Health Report of the Province of Assam, 1921.
1923 —— The season of onset of kala-azar. Ind. Med. Gaz., February, vol. 58, No. 2, pp. 52-56.
1923 —— Some facts in regard to the progress of kala-azar work in the Province of Assam in general and in Sibsagar in particular. Proc. Assam Brit. Med. Assoc., Annual meeting, Haflong, January 7, pp. 10-19.
1924 —— Fourteen years' experience with kala-azar work in Assam. Trans. Roy. Soc. Trop. Med. and Hyg., vol. 18, No. 3, pp. 113-126.
1924 —— The kala-azar transmission problem. (Correspondence.) Ind. Med. Gaz., October, vol. 59, No. 10, p. 529.
1924 —— Kala-azar in Assam. (Lewis and Co.)
1927 —— Some observations on sandflies in Bombay City. Ind. Journ. Med. Res., vol. 14, No. 3, p. 679.
1914 YOUNG, W. (T. C. ?) MCCOMBIE. Segregation and kala azar. A useful measure. Ind. Med. Gaz., vol. 49, No. 8, pp. 301-303.

1908 ZAMBONI. Röntgenterapia nell'anemia splenica infantile. Società medico-chirurgica di Bologna; seduta March 28. Il Policlinico, sezione pratica, vol. 15, p. 528.
1909 —— Ricerche anatoma-patologiche sull'anemia splenica infantile. Rivista di Clinica Pediatrica, No. 10.
1910 —— Osservazioni cliniche e nuove ricerche anatomo-patologiche sull'anemia splenica infantile. Rivista di Clinica Pediatrica, vol. 8, No. 7, pp. 529-585.
1916 ZINCONE, PIETRO. La leishmaniosi interna in Provincia di Caserta. Mal. e Malat. d. Paesi Caldi, January, vol. 7, No. 1, pp. 7-14.

BIBLIOGRAPHY ON BURDWAN FEVER.

(1) ELLIOTT.—Report on Epidemic Remittent and Intermittent Fevers occurring in parts of Burdwan and Nudia Districts, 1863.
(2) Report of the Epidemic Commission, 1864.
(3) GREEN.—Report on the Nature and History of Fever in the Districts of Lower Bengal, 1868.
(4) No. 50, of May 25, 1871, from Ward, Magistrate of Burdwan (referred to in French's paper).
(5) No. 394, of December 30, 1871, from Payne, Officiating Sanitary Commissioner, Bengal. (Supplement to the Calcutta Gazette, January 16, 1872.)
(6) No. 960, of December 27, 1871, from Browne, Inspector-General of Civil Hospitals. (Supplement to the Calcutta Gazette, January 10, 1872.)
(7) GUPTA.—Report on the Burdwan Fever, 1872.
(8) No. 267, of May 16, 1872, from Buckland, Commissioner of Burdwan. (Supplement to Calcutta Gazette, June 5, 1872.) No. 367, of July 6, 1872, from Buckland. (Supplement to Calcutta Gazette, July 24, 1872.)
(9) Report on Sanitary Measures in India, 1872.
(10) JACKSON.—Report on Burdwan Fever, 1873.
(11) LYON.—Burdwan Fever.
(12) VERCHERE.—Extracts from a Diary kept in Burdwan, 1873-74. Indian Medical Gazette, 1873-74.
(13) FRENCH.—Endemic Fever in Lower Bengal, 1874.
(14) FRENCH.—Endemic Fever in Lower Bengal, 1875.
(15) RAY.—Burdwan Fever, 1876.
(16) CHEVERS.—Diseases of India, 1886.
(17) HUNTER.—Imperial Gazetteer of India.
(18) BUCKLAND.—Bengal under the Lieutenant-Governors.
(19) Bengal District Gazetteers (Burdwan and Nudia).
(20) ROGERS.—The Lower Bengal (Burdwan) Epidemic Reviewed, 1897.
(21) ROGERS.—Fevers in the Tropics, 1908.
(22) Transactions of the Bombay Medical Congress, 1909.
(23) Report of the Sanitary Commissioner of Bengal, 1874.
(24) MULLICK.—Nuddia Kahini, 1910.
(25) Indian Medical Gazette, 1871, 1875.

SUBJECT INDEX.

INDEX OF AUTHORS REFERRED TO IN THE TEXT.